



Future Food Systems:
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ABSTRACT BOOK POSTERS

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Exploring consumer attitudes toward plant-based fish analogues for sustainable dietary choices

Marta Appiani¹, Camilla Cattaneo¹, Monica Laureati¹

¹ Sensory & Consumer Science Lab (SCS_Lab), Department of Food, Environmental and Nutritional Sciences (DeFENS), University of Milan, 20133 Milan

Aim:

The food system is facing numerous challenges in advancing environmental sustainability, human health, and animal welfare. A promising solution is the transition towards plant-based diet. Nevertheless, the perceived poor sensory quality of new plant-based products and the resistance to changing eating habits remain significant hurdles for consumers.

Method:

165 consumers (48.5% F; 18-65 years) were clustered based on their attitudes towards food purchase, quality, preparation and consumption using the Food Related Lifestyle questionnaire. The segments of consumers were characterised according to socio-demographic variables, personality traits and frequency of consumption of plant-based alternatives products.

Results:

Three clusters of consumers were identified: 1) The largest and oldest group "Careful consumers" (55.1%) prioritized health aspects and carefully read label information. They placed greater importance on food shopping, price criteria and utilized shopping lists for their purchases. They also showed a higher consumption of plant-based analogues. 2) 'Hedonic consumers' (26.1%) were mainly young consumer inclined to try new food products. Their attention is focused on the sensory aspects and food taste; finally 3) 'Uninvolved consumers' (18.8%) showed less interested in food-related activities and information and are less interested in cooking. They exhibit higher neophobia index and they rank among the least frequent consumers of analogues products.

Conclusion:

Three distinct groups of consumers with significantly different behaviour and attitudes towards the consumption of plant-based analogues were identified. The findings from this study offer valuable insights for the food industry, enabling more targeted communication and educational efforts aimed at promoting sustainable nutrition.

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To eat or not to eat? Edible insects from Slovenian consumers' point of view

Maja Bensa¹, Mojca Jevšnik², Irena Vovk³

¹Research Institute of Faculty of Health Sciences, Faculty of Health Sciences, University of Ljubljana,

²Department of Sanitary Engineering, Faculty of Health Sciences, University of Ljubljana, ³Laboratory for Food Chemistry, National Institute of Chemistry

Aim:

The aim of our study was to investigate Slovenian consumers' opinions and experiences with edible insects through a series of focus group discussions. Protein diversification is striving to make diets more sustainable by including alternative protein sources such as edible insects in consumers' diets. Edible insects have long been a part of the diet in some countries, but the European Union (EU) has only recently approved the following edible insects as a novel food (NF): migratory locust (*Locusta migratoria*), yellow mealworm larva (*Tenebrio molitor*), lesser mealworm larva (*Alphitobius diaperinus*) and house cricket (*Acheta domesticus*). Food safety is an important aspect of NF approvals and NF regulation also states the conditions under which the NF may be used, additional specific labelling requirements and other requirements. Sustainability is important, but such novelties are not always welcomed by the consumers and with many food scandals occurring in the EU in recent years, the importance of open communication with consumers should not be underestimated.

Method:

The information was gathered through a comprehensive consumer food safety investigation, covering topics from shopping to cooking and involving over 30 diverse participants from Slovenia. Focus groups, each consisting of an average of four participants, were conducted both in person and online. Structured discussions lasted approximately 1 to 1.5 hours per session.

Results:

This presentation will highlight consumers' opinions on edible insects, consumers' experiences with consumption of edible insects, concerns regarding incorporation of edible insects into diets and also the conditions under which some consumers would be willing to try eating edible insects.

Conclusion:

The findings of this study can serve as important indicators about the key topics and concerns regarding edible insects that need to be more efficiently addressed and communicated to the consumers. The findings can be useful for regulatory bodies and the food industry producing foods with edible insects in the steps of introducing edible insect food products to the consumers in the EU.

Acknowledgements:

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Valorization of apple pomace in brewing: impact on physicochemical parameters and consumer acceptability of beer

Dr. Nazarena Cela¹, Prof. Marta Bertolino², Alberto Sean Cinzano Marone¹, Chiara Nervo¹, Prof. Luisa Torri¹

¹University Of Gastronomic Sciences, ²University of Turin

Aim:

With the primary goal to promote a shift towards circular economy, also through the recovery of food by-products, this study aimed at exploring the potential application of apple pomace (AP) in the brewing process, mainly focusing on its impact on physicochemical properties, aroma profile and consumers' acceptability of beer.

Method:

Six beer samples, differentiated by apple variety (*Gala*, *Golden* and *Gala Schniga*[®]*SchniCo red*) and AP concentration (60 and 180 g/L) used as beer flavouring, were brewed on laboratory scale (23 L). They were analyzed for colour, pH, alcohol content, polyphenols content and bitterness using the CDR BeerLab[®] system, whereas the aroma profile was assessed using the PEN3 electronic nose. For the sensory evaluation, 98 consumers expressed the overall liking for each sample on a 9-point hedonic scale; then they selected the sensory attributes appropriate in describing the sensory profile of each sample and rated their intensity on a 7-point scale, according to the Rate-All-That-Apply (RATA) method. Finally, consumers expressed their opinion about the appropriateness of using apple pomace in brewing, by using a 7-point scale.

Results:

Physicochemical results highlighted a significant lower pH ($p < 0.05$) in beers with 180 g/L of AP. Consumers' preference was significantly influenced by apple variety ($p < 0.05$), with *Gala* samples being the most preferred by consumers. Compared to samples with 60 g/L of AP, the use of the highest AP concentration (180 g/L) decreased the overall liking, possibly because of a heightened perception of unfamiliar sensory attributes. Principal Component Analysis, performed to better visualize differences between samples according to the perceived intensity of RATA attributes, showed a clear discrimination of samples based on apple variety (60.12% total variance explained). Electronic nose analysis also confirmed the discrimination between samples by apple variety and AP concentration. Furthermore, there was no significant difference ($p > 0.05$) in the consumer perceived appropriateness between using fruit or fruit by-products in brewing, suggesting the consumer interest in sustainable products.

Conclusion:

Therefore, this study provided valuable insights to breweries, encouraging them to implement more sustainable practices, such as the valorization of fruit supply chain by-products through their use in brewing.

European Survey on Fish and Other Seafood Consumption and related Consumer Awareness

Mrs Sofia Ioannidou¹, Anthony Ian Mark Smith¹, Georgios Alaveras¹, Giorgia Zamariola¹

¹European Food Safety Authority

Aim

European Food Safety (EFSA) received a request from European Commission (EC) to provide a scientific report on the frequency of consumption of different species of fish, crustaceans and molluscs and on the effectiveness of MSs advice on their consumption in relation to their contamination with mercury.

Method

A project was designed to assess the consumption frequency of these foods, whether consumers are aware of the presence of contaminants in them and of the existence of consumption advice for limiting consumption due to mercury occurrence and if they consider this advice.

A contract was awarded by EFSA to Ipsos European Public Affairs to run a two-phase survey to collect quantitative data on the consumption of certain fish and other seafood species (with a maximum level of 1.0 mg/kg as established under Regulation (EC) No 1881/2006) among the European population and on Europeans' awareness of advice related to chemical contaminants associated with human health risks and benefits. The identification of the status in MSs, to first understand which MSs have already some advice in place and if they plan to update this and when, was performed. The development of the survey methodology and approach followed, including the development of the questionnaires, and defining the timelines for the surveys.

Results

The first point survey run in July 2023 in 27 EU MSs, Iceland and Norway on adolescents, adults and pregnant women. It included a Food Propensity Questionnaire to collect data on consumption and an Awareness Questionnaire to collect data on awareness of the existence/effectiveness of MSs' advice. The results were shared with the national Authorities to help them decide whether to update their advice. The second point survey is planned for November 2024 only in MSs that plan to update their advice.

Conclusion

The results of both surveys and their analysis will be summarised in an EFSA scientific report, due in December 2025. This will help decreasing uncertainties in the EFSA Opinions where the frequency of fish and seafood consumption is concerned and could eventually propose new measures to achieving a necessary risk/benefit balance of fish and seafood consumption.

Impact of eland meat on the nutritional, sensorial characteristics and consumer's acceptability of meat product

Ali Kozlu¹, Nujamee Ngasakul¹, Sandra Teresita Martín del Campo Barba¹, Tersia Needham¹, Iveta Klojdova¹, Diana Karina Baigts Allende¹

¹Czech University Of Life Sciences Prague

Antelopes are a diverse group of hollow-horned, herbivorous animals primarily found in Africa but also, Asia and North America. Eland bovid species stand out as a highly nutritious option among game meats. It is low in fat, rich in high-quality protein, and a great source of essential nutrients like iron, zinc, and B vitamins, particularly B12, making it an excellent choice for a healthy diet. It has a mild flavor with less gaminess than other wild meats, making it more palatable to a broader audience. It can be considered an excellent alternative for people seeking conventional meats like beef. For this study, the forequarter of a female eland (*Taurotragus oryx*) reared at the CZU Common Eland Research Facilities was used to partially replace beef meat in meatball preparation at varying ratios: a) 20% (A₂₀), b) 40% (A₄₀), c) 60% (A₆₀), d) 100% (A₁₀₀), and control (A₀). The effect of replacement on the nutritional composition, texture, color, and sensory characteristics (appearance, color, flavor, odor, juiciness, tenderness, and overall liking) was evaluated. Our results showed that with higher beef meat replacement, the hardness of the meatballs increased ($p < 0.05$). Lower and higher strength values corresponded to A₀ (3.81 ± 0.18) and A₁₀₀ (5.34 ± 0.10) samples. Regarding color, eland incorporation did not significantly affect meatballs' a* and chroma values. L* and hue values generally increased with substitution, whereas b* values decreased. Regarding the sensorial evaluation, the appearance, color, flavor, tenderness, and overall liking parameters of the eland-meatball samples did not show significant differences compared to the control. However, in the case of odor and juiciness parameters, a significant difference was observed for the A₆₀ and A₁₀₀ samples. Based on our findings, we could partially replace beef meat (up to 60 %) using eland meat without affecting the sensorial characteristics of meatballs for broader healthy meat alternatives.

Dietary intake of plant-based meat consumers versus non-consumers in UK National Diet & Nutrition Survey

Mrs Nicole Neufingerl¹, Laurine Ballintijn², Sander Biesbroek², Anne J Wanders¹

¹Unilever Foods Innovation Center, ²Wageningen University & Research, Division of Human Nutrition & Health

Aim:

Current knowledge on the influence of plant-based meat (PBM) on dietary intakes is mainly based on theoretical modelling studies, while evidence from actual consumption data of PBM consumers is scarce, preventing health authorities from developing clear guidelines on PBM consumption. Therefore, this study aimed to investigate dietary intakes of PBM consumers and compare them to those of non-PBM consumers and to dietary reference values.

Methods:

Cross-sectional dietary intake data of 18–64-year-old PBM consumers (n=101) and non-PBM consumers (n=1845), based on 4-day food diaries, from the UK National Diet & Nutrition Survey Rolling Program (2014-19) were used for the analysis. Independent student t-tests were performed to test for significant differences in food and nutrient intakes between PBM consumers and non-PBM consumers. Average nutrient intakes were compared to the estimated average requirements (EAR), or in case no EAR is established to reference average population intakes (AI), reference nutrient intakes (RNI) or upper limits (UL).

Results:

Average intake of PBM among PBM consumers was 36.5 ± 30.3 g/d. PBM consumers had a higher intake of fruit (+54%, $p < 0.001$), vegetables (+49%, $p < 0.001$), pulses (+42%, $p = 0.04$) and non-alcoholic drinks (+13%, $p = 0.008$), and a lower animal meat intake (-62%, $p < 0.001$) than non-consumers. Nutrient intakes of PBM consumers were more in line with dietary reference values, with higher intakes of fibre (83% vs 62% of AI, $p < 0.001$), polyunsaturated fatty acids (105% vs 96% of AI, $p = 0.005$) and most micronutrients than non-consumers, a lower but adequate intake of protein (180% vs 191% of EAR, $p = 0.001$), and no differences in saturated fatty acid (SFA) (both 122% of UL, $p = 0.82$) and sodium intakes (89% vs 86% of UL, $p = 0.49$). In both groups inadequate intakes of fibre, vitamin D, vitamin E, selenium, and potassium and too high intakes of SFA were found.

Conclusion:

In conclusion, PBM consumers had more favourable food and nutrient intakes than non-consumers, suggesting that PBM can support the transition to a healthier plant-based diet. However, more dietary changes are required to ensure adequate intakes of all nutrients.

Plant-based proteins: barriers and drivers influencing consumption

Ms Mamello Maema¹, Prof Hanli De Beer¹, **Prof Daleen Van Der Merwe**¹

¹Africa Unit for Transdisciplinary Health Research, North-West University

Aim:

Increasing numbers of noncommunicable diseases around the globe have severe consequences for individuals and public health, raising health awareness. The COVID pandemic further increased health awareness, at least with some consumers. Increasing awareness of natural whole foods, and the growing consumption of plant-based proteins substituting animal proteins is associated with increasing health awareness. Additionally, all healthy dietary guidelines promote the consumption of fresh fruits, vegetables and whole grains as part of a balanced diet. The move towards more plant-based diets is also promoted for sustainable reasons. Despite the drive to this transition, consumers experience various barriers, as not all consumers are ready or willing to change their food choices. Thus, this research focused on better understanding the barriers and drivers consumers experience eating plant-based proteins.

Method:

We report on findings from a larger study concerning consumers' barriers and drivers of plant-based protein consumption. Our online survey included purposively selected consumers from South Africa participating in household food purchases (N = 351). Barriers and drivers were determined using a self-compiled 5-point Likert scale and analysed using exploratory factor analysis.

Results:

Our findings bring to light three drivers that support plant-based protein consumption. Respondents on average, expressed agreement with the drivers of the "plant-based protein experience" (mean = 3.70) and "trends (health and sustainability)" (mean = 3.67). This positive response indicates a promising trend towards plant-based protein consumption. However, they were neutral in the area of "knowledge and information" driving their consumption of these proteins (mean = 3.15) (where 1=strongly disagree; 5=strongly agree). On the barrier-side our respondents disagreed on the "social experience" (mean = 2.43) serving as a hindrance to their consumption; however, they were more undecided considering "affordability and availability" (mean = 2.43).

Conclusion:

Respondents in this South African context leaned towards the positive side regarding drivers, also displaying fewer barriers. Affordability and availability might pose a challenge in the categories of some processed plant-based protein foods marketed to mimic the animal-based competitor. Health-promoting campaigns focussing on increasing plant-based protein consumption should focus on education to strengthen consumer knowledge and strategies to improve product information.

Consumer Perception of Alternative Protein Sources: Combining Implicit and Explicit Methodologies

Clara Barnés-Calle¹, Alejandra Bermúdez¹, Oxana Lazo², Maria Costey¹, Lluís Guerrero¹, Phd Elena Fulladosa¹, Anna Claret¹

¹Institute of Agrifood Research and Technology (IRTA), ²Research Center for Applied Biotechnology (CIBA)

Aim:

Consumer behaviour is influenced by many factors including culture, personal experiences and other implicit judgements. The aim of this work was to investigate consumers' perception towards alternative protein sources (vegetal, fungi, algae, insect, and cultured meat) by means of the Theory of Planned Behaviour (TPB) and using the Implicit Association Test (IAT) as a tool to measure hidden or subconscious biases.

Method:

A focus group discussion was organised to qualitatively assess consumer perception regarding animal protein and alternative protein sources and to identify the most prominent beliefs that discern them. After that, a quantitative approach was carried out over 100 participants from two different Spanish cities (Barcelona and Seville), aged between 18 and 50, with gender parity and grouped according to the frequency of substitution of animal protein by alternative proteins in their meals. On one hand, IAT was used to measure the strength of associations between kind of proteins (animal or alternative) and evaluations (positive or negative) to reveal participant's implicit predispositions. On the other hand, TPB was used to examine the effect of several constructs including behavioural, normative and control beliefs on the Behavioural Intention to consume products based on alternative protein sources.

Results:

A more negative implicit attitude toward alternative protein sources than traditional meat was identified in 59% of the participants on the IAT test. The TPB global model showed that Attitude, Perceived Behavioural Control, and Interest for Domain-Specific Innovativeness were the most important predictors of Behavioural Intention. However, participants' implicit attitude had a clear effect on their Behavioural Intention, putting in evidence the importance of subconscious and impulsive motives on consumer decision-making. Moreover, differences regarding Moral Norm and Perceived Behavioural Control were found at city level, which could be attributed to different societal environments between the two regions and indicated that the perception of alternative protein sources could be culturally dependent.

Conclusion:

Implicit beliefs have an important effect on consumer decision-making regarding alternative protein sources. Therefore, combining TPB with IAT is useful to provide a holistic and close-to-reality understanding of consumer's perception on this topic.

The effect of video messages portraying hedonic eating on food consumption in real-life setting

Doctor Terhi Junkkari^{1,2}, Maija Kantola³, Anu Hopia², Harri Luomala³

¹Seinäjäki University Of Applied Sciences, ²University of Turku, ³University of Vaasa

Aim:

Recent studies suggest that hedonic food messages can induce mental simulation of eating and lead to satiation, consequently reducing cravings, food intake and facilitating selection of healthier options. This may turn hedonic food consumption cues into a viable healthy eating approach. This study examines the effect of video messages depicting hedonic eating on food consumption in real-life field experiment.

Method:

The research took place in a SPA-hotel restaurant. Seventy percent of the clientele were elderly pensioners undergoing rehabilitation (control N=972; intervention N=611). The study period was divided into a three-day control phase and a three-day hedonic eating stimulus intervention period. Throughout these phases, the restaurant maintained identical menus in their serving lines, presented in the same order. During the intervention period, participants were exposed to a 6-minute video presenting the consumption of high-calorie savoury foods. Food consumed at the buffet line was weighed, and consumption was measured in grams per customer per day. Main courses offered during the study period were categorized as either healthy or less-healthy options, with vegetable and fish-based dishes considered healthy and those containing meat or sausages classified as less-healthy. Similarly, salads at the buffet line were classified as healthy if they were vegetable-based, while those containing mayonnaise, pasta, or cheese were considered less-healthy.

Results:

The absolute intake (g/customer/day) decreased in all measured dishes and salads (healthy dishes: M=217 vs. M=192; less-healthy dishes: M=61 vs. M=49; healthy salads: M=105 vs. M=90; less-healthy salads: M=37 vs. M=27). However, the relative consumption of healthy salads increased, from 73% to 78%, during the intervention compared to control period.

Conclusion:

Viewing hedonic food imagery reduced the overall consumption of both healthy and less healthy main courses and salads but increased the relative consumption of healthy salads. Our findings suggest that continuous exposure to hedonic food imagery during meals may impede subsequent enjoyment of unhealthy food, but not to same extent healthy food. The findings help marketers and public policy makers in promoting healthy foods.

The role of food safety in food practices of Dutch consumers

Dr Wenjuan Mu¹, Dr Floor van Meer¹, Ir Marc Groenen¹, Ir Wouter Hoenderdaal¹, Ir Wieke van der Vossen-Wijmenga², Dr Esther van Asselt¹

¹Wageningen Food Safety Research, Wageningen University and Research, ²The Netherlands Nutrition Centre (Voedingscentrum)

Aim:

In this study, we investigated different social economic positions (SEP) of Dutch consumers regarding their food safety perception and their food safety practices.

Method:

A literature review has been conducted to get a general understanding on the role of food safety in consumer food practices, which is complemented by quantitative data analysis on two surveys, namely the Dutch National Food Consumption Survey (Dutch National Institute for Health and Environment, 3825 respondents representative of the Dutch population), and the Food Safety Survey with Dutch consumers (Netherlands Nutrition Centre, 2000 respondents representative of the Dutch population) respectively.

Results:

The literature review showed that in general Dutch consumers are not concerned about food safety regardless of their SEP, but a clear perception of food safety seems to be lacking. Furthermore, it showed that low SEP consumers in other EU countries and the United States appeared to be more vulnerable towards food safety risks when conducting food practices. Our preliminary survey analysis results showed that low SEP consumers consumed more raw cold processed meats ($t=2.37$, $p=0.018$) and pork ($t=2.56$, $p=0.011$) which may give rise to related food safety concerns. Although low SEP consumers seemed to be more concerned about food safety risk ($t=2.04$, $p=0.042$), a lower risk of food safety issues was seen in their food safety practices such as food storage ($t=6.34$, $p<0.001$), and hygiene ($t=4.70$, $p<0.001$).

Conclusion:

Although our literature review showed indications that low SEP consumers may be more vulnerable for food safety risk, our preliminary survey results from the Netherlands showed that low SEP consumers reported better food safety practices. By combining findings from literature study and survey analysis, practical insights for the Dutch specific situation regarding food safety perception, safe food practices and dietary choices can be generated, which can be served as a good basis for future development of useful intervention options on improving food safety resilience from a consumer perspective. Differences in SEP of Dutch consumers can be considered when designing future interventions.

Empowering safe food innovation: EFSA's updated multidisciplinary approach for the risk assessment of novel foods

Ermolaos Ververis^{1,2}, Reinhard Ackerl¹, Océane Albert¹, Domenico Azzollini¹, **Dr. Elisa Beneventi**, Esther Garcia Ruiz¹, Wolfgang Gelbmann¹, Maria Glymenaki¹, Eirini Kouloura¹, Laganaro Marcello¹, Maura Magani¹, Leonard Matijevic¹, Vania Mendes¹, Estefania Noriega Fernandez¹, Irene Nuin¹, Gabriela Precup¹, Pablo Rodriguez Fernandez¹, Ruth Roldan Torres¹, Annamaria Rossi¹, Francesco Suriano¹, Emanuela Turla¹, Georges Kass¹, Andrea Germini¹

¹European Food Safety Authority, ²Dept. of Hygiene, Epidemiology and Medical Statistics, School of Medicine, National and Kapodistrian University of Athens

Aim: In compliance with the European Union (EU) regulatory framework, novel foods, those not significantly consumed by humans in the EU before May 15, 1997, must undergo a safety assessment by the European Food Safety Authority (EFSA) prior to potential market authorization. This assessment relies on comprehensive datasets and evidence provided in application dossiers prepared by the respective food business operators. EFSA is currently revising its guidance to applicants on the preparation of such dossiers, and the present work aims to present the main elements of this update.

Method: This revision marks the first comprehensive update of the scientific aspects of EFSA's novel food guidance, upon request of the European Commission. The update aims to integrate recent EU regulatory changes in the field of novel foods, advancements in food research and innovation, and EFSA's experience in novel foods' safety assessment since the centralization of the EU assessment process in January 2018. Additionally, to ensure a practical, meaningful and applicable approach, EFSA actively seeks input from academia, researchers, competent authorities, and industry stakeholders.

Results: The updated guidance focuses on technical and scientific aspects including compositional analysis, identity verification, production methods, toxicological assessment, nutritional analysis, exposure evaluation, and allergenicity testing. It encompasses a wide range of foods and food ingredients, from those derived from cell culture and precision fermentation to plant extracts, novel protein sources, and engineered nanomaterials.

Conclusion: The update underscores EFSA's interdisciplinary approach and highlights EU efforts to ensure safe food innovation for its citizens. Stakeholder engagement is paramount in shaping the guidance, facilitated through various channels including colloquia, webinars, and public consultations. By clarifying further aspects of the EU's regulatory framework for novel foods and EFSA's safety assessment principles, the updated guidance document contributes to fostering innovation within future food systems at the interface of science and progress.

Germination as low-impact process to enhance Basque Beans nutritional properties: protein structure and phytic acid.

Iratxe Olazarán¹, Naiara Andonegi-Mendizabal¹, PhD Shuyana Deba

¹Leartiker Scoop.

Aim:

The consumption of legumes has risen recently due to growing interest in plant-based nutrition trends. Although legumes are an excellent source of essential nutrients and protein, their benefits may be diminished owing to antinutritional factors such as phytic acid. Low-impact technological processes like germination could enhance the nutritional qualities of legumes. This research aims to investigate the germination process in two local Basque varieties of common beans, *Phaseolus vulgaris*, evaluating the impact on protein structure and the reduction of antinutritional factors.

Method:

The methodological approach involves several key steps: (i) Selecting the most promising Basque legume varieties, *Alubia pinta alavesa* and *Alubia de Tolosa*, for valorisation; (ii) Preparing the legumes through hydration and sanitization to ensure optimal germination conditions; (iii) Conducting the germination under controlled environmental conditions (25 ± 2 °C and 90% humidity) over periods of 0, 24, 48, and 72 hours; (iv) Calculate the germination percentage and monitoring pH levels throughout the process; (v) Employing BCA assays for soluble protein quantification and utilizing SDS-PAGE to see changes in protein composition; (vi) Quantifying phytic acid content by colorimetry to assess the reduction of anti-nutritional factors. Statistical analysis was performed by ANOVA followed by Fisher's LSD test.

Results:

The experiment showed stable pH levels throughout the germination process. The germination percentages obtained were 57,6% for *Alubia pinta alavesa* and 67,6% for *Alubia de Tolosa* after 72 hours of incubation. Phytic acid levels decreased by 40% in *Alubia pinta alavesa* and by 10% in *Alubia de Tolosa* from raw to germinated beans over the same period. Protein profiles changed, with a decrease in higher molecular weight proteins and an increase in lower molecular weight proteins, resulting in a more pronounced density of smaller molecular weight bands in germinated beans compared to their ungerminated versions.

Conclusion:

The study confirms that germination effectively improves the nutritional quality of Basque legumes by reducing antinutritional factors and modifying protein structure. Germination led to a significant decrease in phytic acid and transformed protein profiles towards more hydrolysed forms. These findings suggest that germination is a beneficial low-impact technique to enhance the health benefits of legumes.

Can citrus essential oils nano-emulsions be considered food additives? A comprehensive study using *in silico* and *in vitro* approaches.

Madalina Lorena Medeleanu^{1,2}, Antonio Cascajosa-Lira², Ana Belen Cerezo Lopez², Silvia Pichardo Sanchez², Giorgiana Catunescu³, Sonia Ancuta Socaci¹

¹University Of Agricultural Sciences And Veterinary Medicine, Faculty of Food Science and Technology , ²University of Seville, Faculty of Pharmacy, , ³University Of Agricultural Sciences And Veterinary Medicine, Faculty of Agriculture

The analyzed citrus essential oils nano-emulsions are nano-systems obtained by incorporating citrus essential oils (CEOs) in a **surfactant: co-surfactant: water** mixture. EOs are gaining interest due to their properties, being used as food supplements, additives, or alternative medicines and treatments based on natural products. Due to their particle size, nano-emulsions can cross the cell membrane more quickly than other matrices that contain bioactive constituents. The compounds' effects in CEO/CEONES vary greatly depending on their chemical structure, resulting in a wide range related to absorption, distribution, metabolism, and excretion. After being consumed, the human body can produce additional metabolite products of all compounds through Phase I and II reactions. However, the impact of these metabolites is sometimes unclear. Research on these substances contributes to the metabolomics profile by investigating the metabolic alterations that occur during compound transformation. Identifying and quantifying these changes will aid in understanding the full extent of the harmful effects. However, many of these aspects are not fully understood and require further investigation to discuss their possible antagonistic effects. This work aims to establish if the CEO-NEs can be used as a food additive based on their physico-chemical properties. In this case, we have evaluated the bioaccessibility of citrus essential oils nano-emulsions. After completing the digestion process, the digestion products were analyzed for antioxidant activity using the ORAC method. The findings regarding antioxidant activity at three stages of digestion (pre-gastric, post-gastric, and post-intestinal) across the four types of nano-emulsions indicate that *tacle*[®] nano-emulsions exhibited superior retention of antioxidant activity, with a minor increase observed. Conversely, orange nano-emulsions exhibited the most pronounced reduction in antioxidant activity. The volatile profile of the digestion products was also analyzed. Furthermore, we performed an *in-silico* study to obtain a complex and relevant database, which can be successfully used to evaluate CEO constituents regarding their toxicological characterization. Additionally, we performed an *in silico* biotransformation in a simulated intestinal microbiota to confirm *in silico* findings. 187 metabolites were identified from the main clusters (linalyl acetate, sabinene, b-ocimene, g-terpinene, a-terpinene, linalool, and terpineol) using the UHPLC-MS/MS method. Four of them are also present in *in silico* results (**C₁₀H₁₄**, **C₁₀H₁₆O**, **C₁₀H₁₆O₂**, **C₁₆H₂₆O₇**). BeNEs were chosen as the most representative formulation because they have the most complex chemical fingerprint and lowest toxicity. The assays recommended by EFSA were used to assess the toxicity of these NEs (Ames assay for mutagenicity and Micronucleus for genotoxicity). The results show no mutagenicity for the concentrations 0.3-5% and no genotoxicity for 0,001-0.023%. There is a thin line between the chemical properties of the toxicological approach, and it is important to have an overall view of the compounds and whether or not they can be used as food additives.

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Isothermal DNA amplification for a better control of allergenic ingredients in the food value chain

Carla Teixeira², Uxia Rodríguez García¹, Prof Jorge Barros-Velázquez¹, Pilar Calo-Mata¹, Andrey Ipatov², **Dr Marta Prado**¹

¹Universidade De Santiago De Compostela (USC), ²International Iberian Nanotechnology Laboratory (INL)

Aim:

Food allergies are considered by the World Health Organization among the five most important public health concerns. Among most common allergenic ingredients in food production, sesame (*Sesamum indicum*) allergens have been associated with particularly severe reactions with a high risk of anaphylaxis (EFSA, 2004), and it has been related as well with occupational hypersensitivity in bakers and other exposed workers after prolonged exposure to sesame seeds (Keskinen et al., 1991). Therefore, it is very important for control laboratories to have available sensitive and reliable methods for the detection of the presence of sesame in food products, tested and evaluated with different food matrixes.

The main objective of this work was the development of a highly sensitive DNA-based method for the specific detection of sesame in food products.

Method:

Several DNA markers have been evaluated with bioinformatic tools to select the best approach. An isothermal amplification technique has been developed for the specific detection of sesame DNA in food samples, and a qPCR based method has been used as “gold standard” to evaluate the performance criteria of the isothermal amplification based method

Results:

Both *Matk* and ITS1 genes have been selected as appropriate markers for the specific detection of sesame in food, due to their variability among species and copy number per cell was also considered.

A qPCR for the amplification of a fragment of the ITS1 gene has been selected and evaluated for the specific detection of sesame in food. An RPA based method has been developed and the method has been tested with model food products with different concentrations of sesame, and with commercial products with different percentages of sesame in their composition. The performance of the RPA method has been compared with the qPCR method in terms of performance criteria, particularly sensitivity in model food samples and specificity against other common food ingredients and plant material.

Conclusion:

A RPA based method for the specific detection of sesame in food products has been designed and tested with both model and commercial food products, demonstrating a good performance and great potential for decentralized analysis.

Antimicrobial Properties of Fruit Seed Extracts and Potential Application on Vegetable Soup

Doctoral Candidate Yagmur Kucukduman¹, Doctoral Candidate Dilara Nur Dikmetas², Funda Karbancioglu-Guler²

¹Yeditepe University, ²Istanbul Technical University

Aim: In recent years, interest in natural microbial agents instead of synthetic ones to extend the shelf life of foods has increased in food industry in terms of consumer health, food quality and safety, economic evaluation and food sustainability. The aim of this study was to evaluate the antibacterial activity of fruit seeds and their extract against selected gram-positive and gram-negative bacteria and addition of pomegranate seed extract which is the effective extract in vegetable soup to examine the antimicrobial effect against *E.coli* at different storage times and conditions.

Method: Pomegranate, grape and tomato seed extracts minimum inhibitory concentration (MIC) were determined against several bacteria at a concentration of 48-1.5 mg/mL by the agar dilution method. Moreover, grape and pomegranate seed antimicrobial activity were also investigated at 96 mg/mL concentration. Since the most effective extract was found as pomegranate seed, it was added into vegetable soup at two different percentages (%0.6-1.2). Vegetable soups were inoculated with 1×10^4 cfu/ mL *E. coli* and *E.coli* count was examined under two different storage conditions at 8 and 25 °C.

Results: MIC values of fruit seed extracts varied, ranging from 3 to 48 mg/mL for all selected microorganisms. Pomegranate seed was the most effective extract (with the lowest MIC value, 3-12 mg/mL). Pomegranate and grape seed waste provided 99.91%-100% and 51.77%-83.96% bacterial inhibition, respectively. For the vegetable soups stored at 8 °C, there was no statistically significant difference in the number of *E. coli* between control and other samples on a daily basis ($p < 0.05$). Vegetable soup stored at 25 °C containing 1.2% and 0.6% pomegranate seed extract inhibited *E. coli* by 99-100% and 82.63-98.58%, respectively, for all storage periods.

Conclusion: In conclusion, it has been observed that fruit seed waste extracts have antimicrobial effects as a result of *in vitro* studies. The use of waste extract of pomegranate seeds, as a natural antimicrobial in vegetable soup had a positive effect on the shelf life. Fruit seed waste extracts can be used as high added value food additive alternative to synthetic antimicrobials in different food matrixes in terms of both waste recycling and low cost.

Unpacking Dairy Dangers: Biogenic Amines in Lebanese Products - Insights for Food Safety

Dr Iman Dankar¹, Hussein Hassan², Mireille Serhan³

¹Lebanese American University, ²Lebanese American University, ³University of Balamand

Aim:

This study aimed to detect the presence and concentration of six biogenic amines (BAs); histamine, phenylethylamine, cadaverine, spermine, spermidine, and putrescine in different dairy products, investigating the effects of physiological parameters like fat content, pH, ripening time, and production seasons on BA levels. It also compared packed (company-manufactured) versus unpacked (baladi) products, addressing a common practice in Lebanon and other developing countries.

Method:

Based on a food questionnaire survey of 514 participants, 48 samples of the most consumed dairy products were analyzed. These included 12 different packed products from companies and 12 different unpacked (baladi) products. Each was purchased twice during the Summer and Fall seasons. The High-Performance Liquid Chromatography (HPLC) technique was used for BA analysis. The ripening time and pH of each product were recorded.

Results:

Cadaverine was found in all samples, followed by phenylethylamine and spermine. No significant difference was detected in BA levels between full-fat and low-fat products, nor between different seasons. No correlation was found between pH values and BA content. However, total BA levels showed significant differences between packed and unpacked products. All packed samples had slight BA concentration, a range of 9.72 to 39.68 mg/kg, whereas nine unpacked products exceeded the toxicity threshold of 120 mg/kg, indicating non-compliance with safety standards. This is concerning as 65.5% of respondents prefer purchasing baladi products. Additionally, a significant difference was detected between the BA concentration and the type of dairy product analyzed, with Halloumi displaying the highest BA concentrations in both packed (39.68 mg/kg) and unpacked (161 mg/kg) samples, considering it is the top consumed type of cheese according to the survey findings.

Conclusion:

The study provides insights into the BA content in popular dairy products in Lebanon, contributing to food safety and public health. None of the packed dairy samples exceeded the BA toxicity threshold, indicating compliance with safety standards. However, the results from unpacked samples were alarming, highlighting the need for surveillance and awareness to ensure food safety and mitigate health risks associated with dairy product consumption, a situation that is prevalent in other developing countries, emphasizing the importance of this study.

The Allergenic Potential of Common Hops (*Humulus lupulus* L.)

Assoc. Profesor Dorota Piasecka-Kwiatkowska¹, Msc Kinga Blacharska¹

¹Department of Food Biochemistry and Analysis, Poznan University of Life Sciences

Aim:

Common hops (*Humulus lupulus* L.) play a crucial role in brewing, contributing to the bitterness, flavor and aroma of beer. Additionally, they are widely utilized in the supplement market due to their potential health benefits such as antibacterial, anti-inflammatory and antioxidant properties. Despite their widespread use, there has been lack of information on the allergenic properties of common hops. This is particularly noteworthy giving that the closely related *Humulus japonicus* species, contains two protein fractions with proven allergenicity. Therefore, the aim of this study is to investigate the allergenic potential of the hops.

Method:

The study was conducted on leaves, cones and stalks of common hops (*Humulus lupulus* L.) varieties Marynka, Lubelski, Magnum. Samples were lyophilised immediately after harvest. Proteins were isolated through extraction with phosphate-buffered saline (PBS, pH=7,4) and their content was determined using the Bradford method. Immunoreactive properties were assessed using slot blot and Western blotting techniques. Sera from patients allergic to pollen allergens as well as antibodies recognizing the major allergens of birch (Bet v1a), timothy grass (Phl p5b) and artemisia (Art. v.1) were used as recognizing antibodies.

Results:

All analyzed hop samples displayed immunoreactivity, with cones and stems showing higher compared to the leaves. Although there were some differences between varieties, proteins extracted from leaves, cones and stems of common hops (*Humulus lupulus* L.) reacted with antibodies recognizing the major allergens of birch (Bet v1a), artemisia (Art. v.1) and timothy grass (Phl p5b). These extracted proteins react also with IgE present in sera of patients allergic among others to birch, artemisia and timothy grass.

Conclusion:

The obtained results clearly demonstrate that individuals sensitized to pollen allergens, such as birch, timothy grass and artemisia may experience allergic reactions also upon exposure to hops. It suggests a cross-reactivity of hops with various pollen allergens and confirms the allergenic potential of hops.

Antioxidant and antimicrobial extracts from the furoid brown algae *Ericaria selaginoides*: Pretreatment effect.

Doctor Sunuram Ray¹, Doctor Cesar Peteiro², **Doctor Teresa Aymerich**¹

¹Food Safety and Functionality Program. Institute of Agrifood Research and Technology , ²Seaweeds Center, Marine Culture Units "El Bocal" Oceanographic Center of Santander (COST), Spanish Institute of Oceanography (IEO, CSIC)

Aim

Research on the bioactive properties of macroalgae has risen in recent years because there is a great interest in exploring novel food ingredients that may enhance human health and wellbeing. In food application, there is a need to obtain optimized food-grade extracts that could be used as biopreservatives and/or natural antioxidants, among other bioactive properties. While food safety is still a worldwide priority, antioxidants are neutralizing compounds that could minimize damaging of biological processes, thus reducing oxidative stress but also food spoilage. In this sense, alternative technologies such as high hydrostatic pressure (100-1000 MPa), that has been used in the food industry, has emerged as a versatile technology, also to be used for the extraction of active compounds in a quicker and more sustainable way.

This study aimed to assess the effect of high hydrostatic pressure technology (HHP) on the extraction of potential antioxidant and antimicrobials compounds from the furoid brown algae, *Ericaria selaginoides* against *Listeria monocytogenes*.

Method:

Fresh *E. selaginoides* brown macroalgae was collected at the Santander coast, Northwest of Spain. HHP was performed in a Hyperbaric wave 600 pilot equipment. Extraction was performed using a isopropanol-based solvent. Antimicrobial activity was evaluated through determination of minimal inhibitory concentration (MIC) of the crude extracts against *Listeria monocytogenes* CTC1011 in a Bioscreen C microtiter system. Quantification of the antioxidant activity was performed by the determination of the IC₅₀ using DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay.

Results:

The results of the present study showed that HHP did not negatively impact on the antioxidant activity of the crude extracts, The IC₅₀ of HHP-pretreated extracts from *E. selaginoides* was not significantly different from the control extracts, non-HHP pretreated extracts obtained from the fresh algae. On the other hand, HHP extracts significantly improved their antimicrobial activity against *L. monocytogenes* as showed by the 2 to 4-fold decrease in the MIC, thus showing higher potential to combat the food-borne pathogen.

Conclusions:

E. selaginoides crude extracts obtained through HHP pretreatment has optimized antimicrobial activity while kept its antioxidant properties, thus increasing their potential to be applied as biopreservatives and antioxidants to increase food safety and shelf life.

Antioxidant Potential of Pomegranate and Prickly Pear Peel Extracts in Simulated Digestion

Guadalupe Lavado¹, María Pastor¹, Dr Ramon Cava¹

¹University Of Extremadura

Aim:

To assess the impact of incorporating pomegranate peel extract (PomEx) and prickly pear peel extract (PriEx) as natural sources of phenolic compounds (PC) on the generation of 4-hydroxy-nonenal (4-HNE), pentanal, and hexanal in cooked chicken models during *in vitro* digestion.

Method:

Six cooked chicken models were prepared: Control Negative (0 mg/kg NaNO₂), Control Positive (150 mg/kg NaNO₂), PomEx_30 (30 mg/kg TPC and 0 mg/kg NaNO₂), PomEx_300 (300 mg/kg TPC and 0 mg/kg NaNO₂), PriEx_30 (30 mg/kg TPC and 0 mg/kg NaNO₂), PriEx_300 (300 mg/kg TPC and 0 mg/kg NaNO₂). These models underwent *in vitro* gastrointestinal digestion, and the concentrations of 4-HNE, pentanal, and hexanal in the oral, gastric, and intestinal phases were determined. The digests were reacted with 1,3-cyclohexanedione, forming fluorescent derivatives, which were measured using HPLC-FD.

Results:

The three compounds exhibited a similar trend, increasing throughout simulated digestion. Contents of 4-HNE, pentanal, and hexanal were highest in uncured model digests at all digestion phases. Sodium nitrite or PomEx and PriEx addition resulted in a reduction in the formation of 4-HNE, pentanal, and hexanal. Pomegranate peel and prickly pear extracts effectively controlled the formation and accumulation of 4-HNE, pentanal, and hexanal during gastrointestinal digestion. PomEx, at 30 mg PC/kg, showed greater efficacy than PriEx in controlling lipid peroxidation. At equivalent addition levels, PomEx at 30 mg PC/kg had the same antioxidant effect as PriEx at 300 mg PC/kg.

Conclusion:

The removal of nitrite in cooked chicken models resulted in heightened lipid peroxidation, leading to the subsequent formation of 4-HNE, pentanal, and hexanal during simulated digestion. This vulnerability can be mitigated by supplementing with PomEx and PriEx at doses of 30 mg CP/kg and 300 mg CP/kg in cooked chicken models, respectively. Funded by Fondo Europeo de Desarrollo Regional and Consejería de Economía, Ciencia y Agenda Digital (Junta de Extremadura) Una manera de hacer Europa. (Project IB20173).

High Fibre White Bread with Increased Health Benefits using Xylanase

Anna Christian¹, Alison Lovegrove², Ondrej Kosik², Peter Shewry², Ourania Gouseti¹

¹University Of Copenhagen, ²Rothamsted Research

Aim: A low consumption of dietary fibre (DF) is associated with adverse health outcomes, including increased risk for developing type 2 diabetes, cardiovascular diseases, and poorer gut health and motility. Therefore, increasing DF intake is essential for enhancing overall health.

Grain, such as wheat, is a food source that is high in DF. However, when wheat is processed into white flour, a significant amount of the DF is removed. Therefore, increasing the content of DF in white wheat bread, a food consumed worldwide, presents an opportunity to increase DF and promote a healthier diet. This study aims at characterising different wheat flours that are naturally high in DF in the endosperm and examine how adding xylanases to flatbreads made from these high DF flours, affects their digestibility.

Method: Five white wheat flours were examined by analysing their monosaccharide composition and investigating their structures by enzymatic fingerprinting. Upon identifying two flours with the highest and lowest DF content, they were used for making doughs for flat breads.

Xylanase, an enzyme often used in bread making, was mixed into the doughs, and allowed to hydrolyse arabinoxylan (the DF of wheat) at different degrees before baking. DF structures of enzymatically treated flours showed increased content of the monosaccharide xylose, compared to untreated flours. Further, In-vitro digestion, using the INFOGEST 2 method, was used to determine whether addition of xylanase would slow down the rate of digestion, measured as released maltose-equivalents.

Results: The current results indicate that the addition of xylanase for at least 30 minutes to the high DF flat bread can decrease the rate of digestion by about 30%.

Conclusion: Future experiments will explore the effects of varying xylanase concentrations on flatbread digestibility. These findings could be important for the development of high-fibre white bread products with higher consumer acceptance and improved health benefits by utilising wheat variants high in DF in the endosperm.

Preparation of galactooligosaccharides with fucose using β -galactosidase

Anna Macůrková¹, Júlia Vicenová¹, Jiří Štětina¹, **Assoc. Prof. Ladislav Čurda¹**

¹University of Chemistry and Technology, Department of Dairy, Fat and Cosmetics

Aim:

Galactooligosaccharides (GOS) are prebiotics that selectively support the growth of probiotic bacteria and thus positively affect gut health, the immune system, metabolism and can contribute to the prevention of certain diseases. Fucose is richly represented in the oligosaccharides of breast milk or in the polysaccharides of seaweed and has potential for application in functional foods. The aim of the work was to verify the preparation process of oligosaccharides from lactose and fucose by an enzymatic reaction catalyzed by β -galactosidase.

Method:

Lactose and fucose was mixed in ratio weight range from 20:0 to 20:5 in phosphate buffer in pH range 4.5 to 8.0. Enzyme preparation Danisco® Nurica™ was added (1.3 mL per 100 mL of reaction mixture) at 37 °C. The reaction products were analyzed by HPLC on Hypercarb column and ELS detection. GOS were purified on active carbon Norit DX Ultra.

Results:

Maximal amount of fucose was incorporated in to GOS at lactose:fucose ratio 20:4, at pH 5.5 after 60 min reaction. After stepwise extraction of GOS by increased concentration of ethanol GOS preparation containing 38.9 % of GOS was obtained. The GOS preparation consisted mainly of tri- and tetrasaccharides and contained a small amount of di- and pentasaccharides. No monosaccharide were in GOS preparation after purification. The differences in the chromatograms of the reaction mixture with and without fucose show that a smaller amount of oligosaccharides with fucose is formed during the reaction.

Conclusion:

It follows from the obtained results that with the help of β -galactosidase with high transgalactosylation activity, a GOS mixture can be prepared, which contains a smaller amount of oligosaccharides with fucose.

Enhancing nutritional quality of alternative protein foods through nutrient composition, digestibility, and processing

Pinja Pöri¹, Merel Daas², PhD Ever Hernández Olivas³, PhD Sander Biesbroek², PhD André Brodkorb³, PhD Nesli Sözer¹

¹VTT Technical Research Centre Of Finland, ²Wageningen University & Research, ³Teagasc

Aim:

The current food system's unsustainable practices often compromise the nutritiousness of foods, aggravating human health and environmental challenges. The present study combined nutrient composition, digestibility, and food processing of protein ingredients to enhance the nutritional quality of alternative protein foods. Specifically, the focus was on three protein ingredients: faba bean protein, microbial protein, and yellow chlorella concentrate.

Method:

First, individual ingredients underwent nutritional analysis before blending based on optimized nutritional profiles that aimed to resemble meat. Three protein blends were created: (1) 60% faba bean and 40% yellow chlorella, (2) 22.5% faba bean and 77.5% microbial protein, and (3) 13.5% faba bean, 11% yellow chlorella, and 75.5% microbial protein. Meat analogues were produced from the blends by using high-moisture extrusion processing with a laboratory scale twin-screw extruder. Cutting and compression tests were performed by a texture analyser for the extrudates. Finally, to determine *in vitro* protein digestibility, oven cooking was applied and the extrudates were studied before and after cooking.

Results:

The results showed that none of the protein blends could fully resemble the nutritional profile of meat, selenium, and vitamins A, B1 and B3 being most problematic. Nonetheless, combining microbial protein and yellow chlorella in blend 3 created a more beneficial nutritional profile closest to meat. Besides, all blends contained sufficient amounts of protein and met the adult amino acid reference profile of the WHO. From a structural perspective, blend 2 with the highest protein content was the most suitable for extrusion processing although none of the blends formed a clear fibrous structure during extrusion. The yellow algae in blends 1 and 3 affected the process by softening the extrudates' structure, which was also confirmed by instrumental textural analysis. During *in vitro* digestion, cooked extrudates showed a more complex structure, making them slightly less digestible. Overall, protein digestibility resulted higher than 90%. *In vitro* DIAAS showed extrudates as good protein quality with limiting amino acids values higher than 75%.

Conclusion:

This study provides valuable insights into optimizing protein blends for alternative food products, emphasizing nutritional quality, processability and digestibility.

Assessing the mouthfeel of milk alternatives with crystalline dispersed phase using oral tribology

Mr. Philipp Schochat¹, Lina Lepp¹, Prof. Heike P. Karbstein¹, Dr. Nico Leister¹

¹Karlsruhe Institute of Technology (KIT) Food Process Engineering

Aim:

Customers highly value the consistency of milk. The desired mouthfeel can be described with the flow behavior and the friction of milk in the mouth. To improve the mouthfeel of milk alternatives, the friction coefficient and the viscosity in the mouth should be similar to that of milk.

Method:

Milk alternatives were produced with membrane emulsification and different triglycerides as dispersed phase. A 2 wt% whey protein isolate solution was used to stabilize 3.5 % or 30 % of the dispersed phase. By comparing low melting triglycerides (e.g. medium chain triglyceride oil) to high melting triglycerides (e.g. hydrogenated coco-glycerides) the solidity of the dispersed phase was varied. Additionally, each sample was measured at four different temperatures in a range from 5 °C to 50 °C. Rheological measurements with a double gap geometry were carried out to achieve the viscosity over the shear rate (1–1000 1/s). Oral tribology measurements were carried out to achieve friction profiles for each suspoemulsion. The measurements were executed with the one ball on three pins method for different rotational speeds (0.001–100 mm/s).

Results:

Suspoemulsions showed a decreasing dynamic viscosity with increasing temperature, as expected. The differences in solidity (hard, pasty or liquid state) of the dispersed phase caused no differences in viscosity at all temperatures. With increasing solidity of the dispersed phase, the friction coefficient increased. Adapting the solidity of the dispersed phase by replacing liquid oil with blends of plant-based fats mimicked the friction profile of pure butter fat suspoemulsions and cream. While the rheological properties remained unaltered by the change of the triglyceride mixtures, the presented tribology method could detect clear differences.

Conclusion:

The oral tribology of milk and milk alternatives with liquid oil droplets differ significantly from products with solid fat particles. For product development, the dispersed phase content can be used to tune the viscosity and the solidity of the triglyceride phase can adjust the friction values. The study was able to demonstrate how sensitive the measuring device is for investigating suspoemulsions with a crystalline dispersed phase, varied with triglyceride formulation and temperature.

Exploring the metabolic effects of transitioning from high red meat to pulse consumption: De Leguminibus

Gaia Anastasia¹, **Silvia Tagliamonte**¹, Roberto Marotta¹, Paola Vitaglione¹

¹Department of Agricultural Sciences, University of Naples Federico II

Aim:

Global nutritional recommendations advocate for higher consumption of meat alternatives as a protein source due to the adverse health outcomes. Pulses and pulses-based food offer a viable alternative to meat due their proteins and dietary fiber content, however the metabolic advantages and their influence on gut microbiota, remain unclear. This study aims to fill this gap by investigating the effects of replacing red meat with pulses or pulses-based products on cardiovascular risk markers and gut microbiome composition in healthy subjects.

Method:

The study consists in a two-month randomized controlled trial involving 84 healthy subjects characterized by high red meat consumption and low levels of physical activity. Participants will be divided into three intervention groups: one group (28) will maintain their habitual diet (HabD, control), the other groups will receive personalized diets replacing the habitual intake of proteins from red meat with pulses (pulses diet, PulD, 28) or with a combination of pulses-based food products (plant protein diet, PPD, 28). At baseline, after 4 and 8 weeks, fasting blood samples, urine and feces will be collected along with data on anthropometric variables, body composition and blood pressure. Additionally, participants will complete questionnaires to assess diet composition, physical activity, appetite sensations, quality of life, and sleep patterns over the week preceding the visit.

Results:

To date, sixty-two volunteers have signed informed consent: 21 in PulD group (12 M/9 F, average age 35.7±11.7 years, average BMI 24.8±4.1 kg/m²), 22 in PPD group (12 M/10 F, average age 36.4±13.8 years, average BMI 25.7±5.5 kg/ m²) whereas 19 were allocated in HabD group (10 M/9 F, average age 35.4±11.9 years, average BMI 24.7±3.3 kg/ m²).

Conclusion:

We will present the comprehensive study design along with preliminary findings on adherence and clinical parameters, including weight, BMI, body composition, blood pressure, waist circumference, and hip circumference.

Development and *in vitro* validation of a functional fermented chickpea puree

Manuela Flavia Chiacchio¹, Silvia Tagliamonte¹, Angela Pazzanese¹, Francesca De Palma², Martina Coletta², Luisa Cigliano², Giuseppe Blaiotta¹, Paola Vitaglione¹

¹Department of Agricultural Sciences, University of Naples Federico II, ²Department of Biology, University of Naples Federico II

Aim:

Healthy and sustainable diets support increased consumption of plant-protein sources. Legumes possess favourable sustainability credentials and evidence shows that fermentation may improve their nutritional quality. This study aimed at developing and validating *in vitro* an innovative fermented chickpea-based puree with enhanced functional properties compared to unfermented counterpart.

Method:

Fermented purees (FP) containing 10% and 20% w:v of chickpea inoculated with 16 different strains of lactic acid bacteria, along with unfermented purees (CP) were produced and assessed for the content of polyphenols and bioactive peptides (BAPs) along with the total antioxidant capacity (TAC). The most promising FP was selected to undergo *in vitro* digestion (INFOGEST method) to evaluate polyphenol bioaccessibility and TAC release over digestion. Moreover, the anti-inflammatory effect of the FP was investigated in LPS-activated HCT116 cells.

Results:

In the 20% CP and FP, polyphenol concentrations were 1.7- and 1.1-fold higher compared to the 10% counterparts. Five polyphenols were detected in the 20% purees, with synapic acid glucoside being prevalent in CP while pyrogallol in FP. Fermentation notably increased both soluble and direct TAC of purees compared to CP, with *Leuconostoc mesenteroides* and *Lactiplantibacillus plantarum* being the most promising species. Eight BAPs, including Dipeptidyl peptidase-IV (DPP-IV) and Angiotensin Converting Enzyme (ACE) inhibitors were identified, increasing upon the fermentation process. Following digestion, the 20% *Leuconostoc mesenteroides*-treated chickpea puree exhibited a 1.2-fold increase in TAC, while a remarkable 18-fold increase was observed for CP. CP released significantly higher amounts of p-hydroxybenzoic acid, kaempferol-3-O-glucoside, and pyrogallol during digestion compared to undigested puree whereas, FP primarily released pyrogallol showing a bioaccessibility of approximately 140%. FP reduced the inflammation pathways in LPS-treated intestinal cells by reducing TLR4, the degree of activation of the transcription factor NFκB and endoplasmic reticulum stress.

Conclusion:

Fermentation improves the nutritional profile of purees by increasing the concentrations of BAPs and polyphenols. The chickpea puree fermented by *Leuconostoc mesenteroides* showed an increased bioaccessibility of polyphenols and antioxidant activity over the digestion along with anti-inflammatory properties.

Steering in vitro digestibility of lentil-based foods

Dr. Dorine Duijsens¹, Dr. Sarah Verkempinck¹, Prof. Tara Grauwet¹

¹Laboratory Of Food Technology, KU Leuven

Aim: Pulses are increasingly put forward as part of healthy and sustainable diets. Within pulses, macronutrients are enclosed by a cell wall, resilient to cooking followed by mechanical disintegration (*e.g.*, chewing). This encapsulation retards macronutrient digestion, contributing to the low glycemic index of pulses. While pulses are a dietary staple in many regions, their consumption in the western world is limited due to sensory issues and long cooking times. Pulse consumption could be boosted by incorporating them into staple foods (*e.g.*, pasta). Such foods are typically made from raw-milled pulse flours, in which the inherent microstructural organization of pulses is completely disrupted, losing the typical attenuated digestion properties of pulses.

Method: Firstly, the process-structure-digestive function relation of lentils was established. Secondly, these insights were utilized to targetedly process lentils, generating ingredients with different levels of cellular intactness. Next to isolated cotyledon cell powders, whole precooked lentil powders were employed to avoid the generation of fiber-rich waste streams (*e.g.*, hulls). Ingredients were evaluated in terms of *in vitro* macronutrient digestion kinetics in a model food system (*i.e.*, heat-treated suspension). Thirdly, cellular lentil ingredients were incorporated into lentil-based pasta as a real food product, and evaluated in terms of cooking properties and *in vitro* starch and protein digestion behavior.

Results: Compared to a raw-milled lentil flour, cellular ingredients showed significant attenuation of starch and protein digestion, affected by different processing parameters (*e.g.*, precooking time and drying method). Upon incorporation into a pasta, cellular ingredient incorporation showed little effect on the cooking properties. The cellular structure was maintained during pasta preparation, causing a 16-25% reduction of *in vitro* starch digestion compared to a pasta made solely from raw-milled lentil flour. In contrast, the level of proteolysis was not significantly affected.

Conclusion: In conclusion, lentil ingredients and pastas with (partial) cellular intactness showed improved nutritional properties characterized by a slowed starch digestion but high level of protein digestibility. These ingredients can be utilized for developing innovative pulse-based foods with targeted digestion kinetics, also for specific population groups requiring adapted nutrition.

Polysaccharide coated nanoliposomes improve polyphenol intestinal transepithelial transport

Xiangnan Meng¹, Dr. Christos Fryganas¹, Prof. Vincenzo Fogliano¹, Dr. Tamara Hoppenbrouwers^{1,2}

¹Food Quality and Design, Wageningen University and Research, ²Wageningen Food and Biobased Research, Wageningen University and Research

Aim:

Encapsulation is largely used to protect bioactive compounds during food product manufacturing and storage. Hesperetin (HST) is a potential anti-inflammatory flavanone, but its bioavailability and biological activity are partly limited because of poor water solubility. We chose HST as an example of hydrophobic compounds, to investigate the improvement of the bioavailability and anti-inflammatory properties by using different delivery systems: maltodextrin (MD), β -cyclodextrin (CD), nanoliposomes (NL), chitosan coated nanoliposomes (CH-NL), and carrageenan-chitosan coated nanoliposomes (CGN-CH-NL).

Method:

Spray-drying and freeze-drying were applied to encapsulate HST in MD and CD. NL were made by the thin film hydration method followed by sonication. The coated NL were prepared by drop-wise polysaccharide solutions. The physical properties, phenolic metabolites of different delivery systems, release profile during *in vitro* digestion, and cell transport were also investigated. A direct Caco-2 and THP-1 cells coculture model to test the anti-inflammatory effect of delivery systems was used. Target cytokines were measured by ELISA, and the metabolites produced by the coculture model were analyzed by LC-qTOF-MS.

Results:

Physical characterization showed the successful preparation of delivery systems. Fourier-transform infrared spectroscopy showed CH and CGN properly coated the NL vesicles. After *in vitro* digestion, 76% of delivered HST was effectively kept in the CGN-CH-NL, whereas delivery systems such as MD and CD retained in the carriers only 30% and 66%, respectively. In only the Caco-2 cell model, CGN-CH-NL showed the highest transported HST through the intestinal epithelium and maintained its a threefold increase compared to free HST. In only the THP-1 cell model and the coculture model, CGN-CH-NL-HST significantly decreased the IL-8 production.

Conclusion:

The development of CGN-CH-NL in this study allowed the controlled release of HST in a GI tract simulation model and improved cellular uptake after *in vitro* digestion. Moreover, it also has a potential anti-inflammatory effect in decreasing the secretion of proinflammation cytokines IL-8. The strategy of designing polysaccharide coated nanoliposomes can be a promising tool for promoting HST transepithelial transport in the small intestine and contribute to the anti-inflammatory action of dietary compounds.

Avocado Residual Pulp Extract in Enhancing Artisanal Ham: Insights from *in vitro* Digestion Studies

Maria Elena Sosa-Morales¹, Nill Campos-González¹, Julián A. Gómez-Salazar¹, Abel Cerón-García¹, Fabiola León-Galván¹, Luz E. Casados-Vázquez¹, Sonia Sáyago-Ayerdi², Everardo Mares-Mares³
¹Universidad de Guanajuato, ²Tecnológico Nacional de México, ³Tecnológico Nacional de México

Aim:

In vitro digestion techniques provide an alternative approach to investigate the bioaccessibility of bioactive compounds, previous to animal or human studies. Currently, there is limited research demonstrating how meat-products can influence the bioaccessibility of bioactive compounds derived from agri-industrial by-products. avocado residual pulp (ARP) is generated from the oil avocado industry and still has good enough biocompounds, with potential to be revalorized. The aim of this study is to assess the bioaccessibility of phenolic compounds from optimized extract of avocado residual pulp (ARP) incorporated into baked pork ham.

Method:

Extract of ARP was obtained by microwave-assisted extraction at optimized conditions. Control artisanal ham (C, without addition of extract) and ham added with ARP extract at 1.5% (T-1.5) were submitted to an *in vitro* gastrointestinal digestion. Samples underwent enzymatic hydrolysis mimicking both gastric (GD) and intestinal digestion (ID). Subsequently, the soluble (SIF) and insoluble indigestible fractions (IIF) were retrieved. The total phenolic compounds content (PCs) associated with each stage and phase of digestion were assessed with Folin-Ciocalteu reagent to determine the bioaccessibility percentages for each sample.

Results: PCs content increased as the sample underwent ID for both samples, with T-1.5 showing the highest content of these compounds. The quantification of PCs in the SIF was minimal, and in the case of the IIF, they were not detected (n.d). The PCs in the SIF was higher in the ham with 1.5% ARP in comparison to control ($p \leq 0.05$). The bioaccessibility values were $98.48 \pm 0.57\%$ for the control ham and $96.16 \pm 0.08\%$ for T-1.5. Both hams had high bioaccessibility percentages, indicating that the enzymes used can hydrolyze the components of the meat matrix, allowing the phenolic compounds that may be bound to macromolecules such as carbohydrates and proteins to be released. This behavior suggests that most phenolic compounds are available to be assimilated by the body and exert their beneficial effects.

Conclusion:

Baked ham matrix did not retain the phenolic compounds coming from the added ARP extract, making them easily bioaccessible. The use of the extract of this residual at 1.5% has potential to enhance the quality of a meat product.

Sustainable protein blends for infant nutrition. Balancing health and sustainability.

Dr Maria Jose Bernal¹, Dr. Michelle Klerks¹, Marie Breer¹, Dr. Luisma Sanchez-Siles¹

¹Hero Institute for Nutrition, Hero Group

Aim: During weaning the protein quality is key for supporting adequate growth. Although animal proteins have better quality as compared to plant proteins, the last recommendations for sustainable and healthy diets Planetary Health Diet encourage a decreased ratio of animal:plant protein consumption. Achieving high protein quality in baby foods with both types of proteins poses a challenge, thus a balance between both is of importance. This study aimed to evaluate the amino acid (AA) profiles of more sustainable commercial infant meals containing animal-plant protein blends (APPBs).

Method: Four APPBs were designed and included in infant meal recipes. In total six infant meals were analyzed, including the four APPBs recipes with varying combinations of meat, legumes, and cereals as main protein sources (chicken 4-5%, beef 4%, lentils 9-11.5%, white beans 7-8%, cereals 5-12%) and two standard recipes with meat as protein source (chicken 9% and beef 8%). Eleven essential AA for infants were assessed using High-Performance Liquid Chromatography (HPLC). The AA scoring patterns were evaluated using the FAO standards. Based on the portion sizes, the total AA intake was estimated and compared against the Recommended Dietary Allowance (RDA) for infants and toddlers aged 7-36 months. Unpaired t-tests were conducted to test for differences between standard and APPBs recipes.

Results: Adequate and similar AA levels were observed in both APPBs and standard recipes ($p > 0.05$). No significant differences were found in the RDA (%) of AA per portion/day between the standard and APPB recipes, with adequacy observed in all recipes (mean 54% of RDA).

Conclusion: The partial substitution of meat with legumes as traditional protein source in commercial infant meals did not compromise the essential AA intake. This finding indicates that animal-plant protein blend meals can be part of a balanced diet for infants and toddlers. This approach not only promotes the consumption of legumes but also aligns with the principles of the Planetary Health Diet, supporting sustainable dietary choices.

How can gastronomy promote healthy ageing? Exploring its potential in the prevention of Cognitive Decline.

Olaia Estrada¹, Jara Domper Jiménez^{1,2}, Miguel Ruiz-Canela^{3,4}, Usune Etxeberria^{1,2}, Maria Arrizabalaga^{1,2}, Pablo Martínez-Lage⁵, Mirian Ecay-Torres⁵, Mikel Tainta⁵, Miren Altuna⁵, Naia Ros⁶, Goretti Soroa⁶, Imanol Reparaz-Escudero⁷, Mikel L. Saéz de Asteasu^{7,8}, Mikel Izquierdo⁷, Elena Alberdi⁹, Estibaliz Capetillo-Zarate^{9,10}, Maider Mateo-Abad¹¹, Javier Mar Medina¹¹

¹BCC Innovation, Technology Center in Gastronomy, Basque Culinary Center, ²Basque Culinary Center, Faculty of Gastronomic Sciences, Mondragon Unibertsitatea, ³Department of Preventive Medicine and Public Health, University of Navarra, IdiSNA, ⁴Consorcio Centro de Investigaciones Biomédicas en Red (CIBEROBn), Institute of Health Carlos III (ISCIII), ⁵Center for Research and Advanced Therapies, CITA-alzhéimer Foundation, ⁶University of the Basque Country UPV/EHU, ⁷Navarrabiomed, Hospital Universitario de Navarra (HUN) – Universidad Pública de Navarra (UPNA), IdiSNA, ⁸CIBER of Frailty and Healthy Aging (CIBERFES), Instituto de Salud Carlos III, ⁹Achucarro Basque Center for Neuroscience, and Department of Neurosciences, University of the Basque Country UPV/EHU, ¹⁰IKERBASQUE, Basque Foundation for Science, ¹¹Biodonostia Health Research Institute

Aim:

One in three cases of dementia could be prevented by modifying risk factors, including those related to food behaviours. Food-related behaviors, including cooking practices, are considered relevant factors in adopting of a healthy dietary pattern, and therefore in promoting of a healthy ageing. The study aims to assess the impact of a nutritional and culinary intervention to modify food-related behaviors as part of a multimodal approach to preventing cognitive decline.

Method:

The CITA GO-ON study (NCT04840030) is a randomised, controlled, non-pharmacological, two-year efficacy trial of a multimodal intervention to prevent cognitive decline. Participants aged 60-85 at risk of cognitive decline were allocated (1:1) to receive Regular Health Advice (RHA) or a lifestyle-based Multidomain Intervention (MD-Int). The MD-Int consisted of: 1) cognitive training, 2) socio-emotional skills, 3) multicomponent Vivifrail tailored exercise program, 4) cardiovascular risk factor monitoring, and 5) Nutritional and Culinary Intervention (NCI). The NCI included eight face-to-face sessions, educational materials, and direct contact with dietitians and chefs, while the RHA group was given general healthy eating advice. The 14-item Mediterranean Diet Adherence Screener (MEDAS) questionnaire was used to assess adherence to the Mediterranean diet (MedDiet) every four months during the MD-Int. Mixed-effects linear models were used to assess the effect of the NCI within and between groups on MEDAS score. $p < 0.05$ level was considered as statistically significant.

Results:

First-intervention year was completed by 214 participants (115 RHA and 99 MD-Int). Participants in both groups showed a statistically significant improvement in MedDiet adherence over the first year. Although non-significant between-group differences were observed (0.31; 95%CI, -0.2 to 0.9), the mean change in the MD-Int group was slightly higher (1.71; 95%CI, 1.3 to 2.1) than in the RHA group (1.39; 95%CI, 1.0 to 1.8).

Conclusion:

Culinary-nutrition interventions can be successful in promoting healthy eating habits. Further results from the MD-Int are expected to explore the full capacity of these interventions to foster healthier habits that contribute to healthy ageing and the prevention of dementia. Specifically, the initial and concluding assessments of home cooking habits and comprehensive dietary information will be conducted to ascertain the impact of the NCI.

Gut microbiota network analysis to define the “Metabolically Healthy Obese microbiome”: An AI4Food approach

Blanca Lacruz - Pleguezuelos^{1,2}, Laura Marcos-Zambrano¹, Adrián Martín-Segura^{1,3}, Alberto Diaz-Ruiz³, Guadalupe X. Bazán⁴, Isabel Espinosa-Salinas⁴, Lara P. Fernández⁵, Ana Ramírez de Molina^{4,5}, Vera Pancaldi^{6,7}, Enrique Carrillo de Santa Pau¹

¹Computational Biology Group, Precision Nutrition and Cancer Research Program, IMDEA Food Institute (CEI UAM+CSIC), ²Molecular Biosciences Program, UAM Doctoral School, Autonomous University of Madrid, ³Laboratory of Cellular and Molecular Gerontology, Precision Nutrition and Aging Program, IMDEA Food Institute (CEI UAM+CSIC), ⁴GENYAL Platform, IMDEA Food Institute (CEI UAM+CSIC), ⁵Molecular Oncology Group, IMDEA Food Institute (CEI UAM+CSIC), ⁶Centre de Recherches en Cancérologie de Toulouse, CRCT, Université de Toulouse, Inserm, CNRS, ⁷Barcelona Supercomputing Center

Aim:

Obesity is a chronic disease placing a heavy burden in public health systems due to its lifelong consequences on health and wellbeing. Currently, there is discussion on the existence of a phenotype exempt of these risks, called “Metabolically Healthy Obesity” (MHO). As the ensemble of microbes residing in the gut, or gut microbiota (GM), is relevant in physiological processes related to obesity, and given its potential for personalised interventions, we aim to define whether a “MHO microbiome” exists.

Method:

We collected anthropometric, biochemical (lipidic, glycemia, inflammation) and GM whole genome shotgun data for 98 participants. Following guidelines from the BioShare-EU Healthy Obese Project (10.1186/1472-6823-14-9), we classified subjects as MHO (n = 14), metabolically unhealthy obese (MUO, n = 31), metabolically healthy non-obese (MHNO, n = 24) or metabolically unhealthy non-obese (MUNO, n = 29) based on the presence of individual components for metabolic syndrome: increased fasting blood glucose, hypertriglyceridemia, elevated blood pressure or decreased HDL cholesterol. We evaluated their GM through alpha- and beta-diversity measurements and used co-occurrence networks to study its structure. Networks were built with the SPIEC-EASI method, adapted to the statistical challenges of GM data, and analysed through calculation of network topological properties, resistance against attacks, and analysis of rich-club ordering, a phenomenon where the dominant microbes in the network form tightly connected communities.

Results:

The biochemical and anthropometric values showed differences between groups in body composition, inflammation (TNF- α) and insulin levels, as well as the homeostatic model assessment (HOMA) index. No differences regarding GM diversity were found. Network stability analysis shows the MHNO microbiome is the most robust against random attacks, while MUO subjects seem to have the least resistant GM structure overall. GM networks from MUO and MHO subjects show rich-club ordering, while networks from non-obese groups show mixed behaviours.

Conclusion:

Our analyses show structural differences mainly between obese and non-obese groups. Therefore, we do not have evidence to support the existence of a “MHO microbiome” as such, in line with clinical research that questions the relevance of this phenotype.

Nutrient and food group differences in six-month-olds based on weaning method: Dastatuz trial preliminary results

Jone Guenetxea-Gorostiza^{1,2}, Leire Mazquiarán-Bergera¹, Iratxe Urkia-Susin³, Olaia Martinez^{3,4}, Leire Cantero Ruiz de Eguino⁴, **Silvia Matias Ibañez**⁴, Edurne Maiz^{2,5}, Diego Rada-Fernandez de Jauregui^{1,4}
¹Department of Preventive Medicine and Public Health, University of the Basque Country (UPV/EHU),
²Mental Health and Psychiatric Care Group, Biogipuzkoa Health Research Institute, ³Nutrition and Food Safety group, Bioaraba Health Research Institute, ⁴GLUTEN3S Research Group, Department of Pharmacy and Food Science, University of the Basque Country (UPV/EHU), ⁵Psychology and Research Methodology Department, University of the Basque Country (UPV/EHU)

Aim:

Disparities in nutrient intake and food groups offer between weaning methods in six-month-old babies: preliminary results from the Dastatuz trial

Method:

Data of 27 babies at 6 months of age was analysed using 24-hour dietary recalls. Nutritional intake was assessed using the BEDCA and CIQUAL databases and the R software. Categorization of foods into group foods was also performed using both databases. In these preliminary analyses only complementary food was considered, and thus information regarding nutrient intake or offer from breastfeeding or bottle feeding is not included.

Results:

BLW group took 20 % of the energy needs through complementary feeding, while PLW took 38 %, consequently we assumed that those percentage of the recommendations should be accomplished for each nutrient. Nutrient consumption disparities were found between groups, although those differences were not statistically significant. BLW reached the 20 % of EFSA recommendation in protein and sodium, while PLW group reached in protein, sodium and calcium. Neither group reached the stated threshold for fat and iron. In all the nutrients, intake was lower in the BLW group. Additionally, BLW group consumed 8.97 % more of "Vegetables and vegetable products" and 5.06 % more of "Fats and oils". PLW group consumed 3.28 % more "Grains and grain products", 5.26 % more "Milk and milk products" and 1.8 % more of "Fish, molluscs, reptiles, crustaceans and products".

Conclusion:

Nutritional intake from complementary feeding differs according to the chosen weaning method. These differences might come from the relative offer of food groups, which is also different between weaning methods. To assess the actual impact within the whole diet a broader sample should be used, including information from breastfeeding. It also needs to be explored whether the differences seen disappear or are maintained with age.

Bio-calcium from salmon frame prevented bone loss in ovariectomy-induced osteoporosis rats

Prof.Dr. Soottawat Benjakul¹, Dr. Acharaporn Issuriya², Dr. Watcharapol Suyapoh³, Dr. Krisana Nilsuwan¹, **Dr. Jirakrit Saetang¹**

¹International Center of Excellence in Seafood Science and Innovation, Faculty of Agro-Industry, Prince of Songkla University, ²Division of Health and Applied Sciences, Faculty of Science, Prince of Songkla University, ³Department of Veterinary Science, Faculty of Veterinary Science, Prince of Songkla University

Aim:

Postmenopausal women are considered as the high susceptibility group to osteoporosis as witnessed by low bone density and strength, which may lead to bone fracture. Fish Bio-calcium (Bio-Cal) has been recognized as a potential supplement. It is derived from fish processing by-products and it shows high nutritional value, bioavailability, and absorption. Although the characteristics of Bio-Cal have been well documented, the role of Bio-Cal on the attenuation of bone loss in osteoporosis has not been revealed. Therefore, this study aimed to evaluate the effect of salmon-derived Bio-Cal (sBio-Cal) on bone density in ovariectomy-induced osteoporosis rats.

Method:

Sprague-Dawley female rats (n = 20) were subjected to ovariectomy (OVX) or sham surgery. After surgery, OVX rats were divided into 3 groups of 5 individuals each. Those included ovariectomized rats untreated (Negative); OVX rats treated with calcium carbonate (OVX-CaCO₃); and OVX rats treated with salmon Bio-Cal (OVX-sBio-Cal). After 10 weeks of treatment, femur and tibia were collected, and used for analysis, including bone area/total area (BA/TA), trabecular thickness, and cortical bone thickness. Tartrate-resistant acid phosphatase (TRAP) staining assay was used for osteoclast identification.

Results:

Bone histomorphometric analysis showed that BA/TA, trabecular thickness, and cortical bone thickness of femur and tibia from sham, OVX-CaCO₃, and OVX-sBio-Cal were increased compared with the negative OVX (p<0.05). There was no difference in BA/TA between sham and all calcium-treated groups. TRAP staining revealed a higher number of osteoclasts in negative OVX, compared to sham, OVX-CaCO₃, and OVX-sBio-Cal (p<0.05). No difference in osteoclast number was observed between OVX-CaCO₃ and OVX-sBio-Cal (p>0.05) while sham showed higher levels of osteoclast than calcium-treated groups (p<0.05).

Conclusion:

This study revealed that sBio-Cal could be an alternative to calcium carbonate with the same prevention effect of bone loss to prevent osteoporosis in an animal model.

Analysis of commercial yogurts texture connected to dysphagia consumer needs

PhD Esther Sanmartin¹, Ana B. Naranjo¹, Raquel Llorente¹, Irene Peral¹

¹AZTI, Food Research, Basque Research and Technology Alliance (BRTA)

Aim:

Yogurt is a product consumed all over the world, known for its health benefits. Its moderate viscosity and firmness make it particularly well-suited for individuals managing dysphagia. Among other benefits, it serves as a facilitator in medication administration. Yogurts are highly valued by dysphagia consumers for their palatability, and the market offers a vast array of choices, however, the textural characteristics relevant to dysphagia, have not been fully elucidated. Incorporating new data on textural characterization in this widely accessible product, will enable a more targeted and safer product selection based on dysphagia consumer needs.

Method:

A selection of yogurts taken from the market was analysed. Different categories of yogurts were selected based on the classification used by manufacturers. Rheological characterisation of the yogurts was conducted using Discovery HR-10 rheometer (TA instruments Ltd., UK). Moreover, textural characteristics were evaluated using a texture analyser TA.XT2i (Stable Micro Systems Ltd., UK).

Results:

The rheological behavior of the yogurts was observed to display shear thinning behavior across all tested yogurts and categories. When viscosity was measured at a shear rate of 50 s⁻¹, most of the tested yogurts fell into the honey-like classification according to the National Dysphagia Diet (NDD) thickening levels. Furthermore, it was found that categories with higher viscosity, exhibited greater heterogeneity in the tested rheological and textural parameters (e.g. adhesiveness).

Conclusion:

The present study showed the heterogeneity in texture behavior that exists in the market yogurts, even within the same commercial category. Identifying these dysphagia-related properties in commercial yogurts would enable a more informed choice and thus, a products selection more aligned with dysphagia consumer needs.

Antioxidant and antidiabetic properties of tomato-based functional bars

Phd Student Elena Tomassi¹, PhD Morena Gabriele¹, PhD Laura Pucci¹

¹Institute of Agricultural Biology and Biotechnology-CNR

Aim:

With a view to sustainable nutrition, increasingly focused on the use of plant proteins over animal proteins with high environmental impact, Middle East Technical University (METU) produced, as part of the European FunTomP (Functionalized Tomato Products)-PRIMA project, tomato and olive powder-based bars that differ in the type of protein added, pea or rubisco, and by the microwave (MW) and oven cooking method. This work analyses the antioxidant and bio-functional properties of four bar samples selected based on lycopene content, textural properties (i.e., hardness, gumminess, chewiness), organoleptic properties (sensorial properties), sustainable and optimized processing (by multiple response surface methodology in drying), and minimum amount of raw material usage.

Method:

The content of the bioactive molecules (polyphenols and flavonoids) and the antioxidant activities (DPPH, ORAC and FRAP assays) of water and ethanolic (50% water v/v) bars extracts were evaluated. The antidiabetic properties of bars were determined as α -amylase and dipeptidyl-peptidase IV (DPP IV) inhibitory activity.

Results:

A higher content of polyphenols and flavonoids was detected in the bars with rubisco than those with pea protein. Regarding the cooking method, only bars with pea protein showed higher polyphenol levels after oven cooking than after MW. Antioxidant power by DPPH was significantly higher in bars with rubisco cooked with MW than in the others with pea protein ($p < 0.05$). ORAC assay didn't show any significant differences for cooking methods, although the bars with rubisco cooked by MW had higher values than the pea ones ($p < 0.05$); this result was confirmed by FRAP assay. Moreover, FRAP analysis highlighted an improvement of the antioxidant power in bars with rubisco cooked by oven compared to the bar with pea protein cooked by MW ($p < 0.005$). Finally, findings on the α -amylase didn't show any inhibition activity. Otherwise, a significant inhibition of the DPP IV enzyme was observed in bars containing rubisco ($44.3 \pm 3.9\%$ MW and $46.8 \pm 1.2\%$ oven) compared to those with pea protein ($16.3 \pm 2.8\%$ MW and $14.1 \pm 0.4\%$ oven), with no difference between cooking methods.

Conclusions:

In conclusion, the formulated bars showed antioxidant and antidiabetic activity, highlighting rubisco as an alternative protein with strong bio-functional power.

Milk-derived biopeptides: A potential strategy for hypertension treatment

Cindy Stephanny Alvarado Molina¹, Bertha Viviana Ruales Guzmán²

¹Universidad Nacional de Colombia, Department of Chemical and Environmental Engineering,

²Universidad Nacional de Colombia, Instituto de Ciencia y Tecnología de Alimentos

Aim:

One type of treatment used to reduce high blood pressure and heart failure is angiotensin-converting enzyme inhibitors. They act on the activity of the enzyme responsible for converting angiotensin I into angiotensin II, which causes narrowing of blood vessels. Due to the importance of the above, searches have been encouraged to identify alternative sources to obtain these proteins, such as animal milk. Therefore, the objective of this work was to analyze the articles that have investigated angiotensin-converting enzyme inhibitory peptides extracted from animal milk as well as, the scientific papers that have performed clinical trials to test the action of these biopeptides in living beings.

Method:

This research was achieved through a systematic review of the literature in order to identify articles that referred to the influence of milk biopeptides on hypertension, considering the SCOPUS bibliographic database.

Results:

Three main findings were discovered in this investigation. First, studies used casein hydrolysates and animal matrices as sources to extract the biopeptides. In works that used the animal matrix, 86% used proteins of bovine origin, including cows, buffaloes, and yaks, while the remaining percentage were reports of milk from goats and sheep. Second, the main biopeptides examined in these articles were Ile-Pro-Pro and Val-Pro-Pro, with percentages of 25% and 19%, respectively, of the total articles reviewed. Finally, 42% of the entire papers analyzed executed clinical studies on the effects of biopeptides, finding a positive effect on blood pressure, of which 47% were carried out in rats and the difference in humans with mild hypertension.

Conclusion:

According to the results of this systematic literature review, milk proteins are a natural source of bioactive substances that may be useful in the treatment of hypertension. However, in order to assess its impact more accurately on human health, it would be pertinent to conduct long-term studies with a sizable sample of participants comparing different types of animal matrices. These investigations may yield important data that will help evaluate whether it is feasible to use these bioactive peptides in the creation of medicines to treat the hypertension.

Rational design of cereals for seniors: Fabrication, oral processing, palatability and digestibility of cereal prototypes

Miss Leehen Mashiah¹, Mr Omer Medini², Miss Andrea Araiza Calahorra³, Miss Josefina Skaret⁴, Msr Carmit Shani Levi¹, Miss Lotti Egger⁵, Mr Reto Portmann⁵, Ms Paula Varela⁴, Ms Anwasha Sarkar³, Mr Uri Lesmes¹

¹Technion, ²The International Beer Breweries LTD, ³Food Colloids and Bioprocessing Group, School of Food Science and Nutrition, University of Leeds, ⁴Nofima, ⁵Institute for Food Science, Agroscope Switzerland

Aim:

The rise in life expectancies and gap between life span and health span necessitate innovative interventions and food solutions to secure healthy aging. This study, conducted within the EAT4AGE JPI-HDHL project, focuses on the rational design and evaluation of the digestibility co-extruded plant-based functional cereals tailored for older adults. The product, fortified with two bioactive moieties, Maca root powder and Olive leaf extract, aims to address nutritional gaps identified in older adults, with a particular emphasis on high-quality and highly digestible proteins.

Method:

First, nutritional design of a palatable vegan food alternative for seniors was performed and a co-extrusion process was optimized to fabricate three prototypes of ready-to-eat puffed cereal with oil-based filling: without functional ingredients, and with one or two bioactives added. In turn, cereal prototypes were subjected to sensorial testes in two cohorts one of community dwelling seniors (n=21, age>65) and another panel of trained professionals (n=10). Concomitantly, digestive breakdown of the products was studied using for its tribology and *in vitro* digestibility in seniors using SDS-PAGE, LC-MS/MS proteomics and *in vitro* digestible indispensable amino acid score (DIAAS) determinations.

Results:

The cereals have rich macronutrient profile exceeding 12% (w/w) protein, 20% (w/w) fat, and low sugar (<5% w/w), surpassing commercially available products. Texture analysis indicates supplementation improved product expected hardness whilst presence of coalesced oil-driven tribofilm reduced friction and provided oral comfort. The first consumer panel confirmed high palatability regarding bolus formation, low mouth pain, good texture, flavor, and overall acceptability. The trained sensory panel described the sensory attributes resulting from the fortification. Lastly, the product digestion was explored through an age-tailored *in vitro* digestion model. This consistently demonstrated the prototypes have high protein digestibility surpassing 80% and lysine as the limiting amino acid in DIAAS scoring.

Conclusion:

This study highlights the potential of designing palatable foods for healthy aging with potential to harness co-extrusion to fabricate plant-based products that are age-tailored and exhibit high protein digestibility. This work highlights the potential to develop healthier food choices through innovative research tools for studying food digestion and understanding of consumer needs, particularly as it pertains to seniors.

Nudging fragile population towards sustainable food choices

Chiara Biggi¹, Davide Menozzi¹, Audrey Cavalieri¹, Sara Ottonello¹, Riccardo Vecchio², Silvia Sapio², Cristina Mora¹

¹Università degli Studi di Parma, ²University of Naples Federico II

Aim:

Governments have implemented various measures aimed at fostering healthier and more sustainable eating habits among the population. These measures encompass various interventions, ranging from soft instruments such as educational campaigns and disseminating information to influence people's perceptions and attitudes toward food choices, to harder methods like taxation and subsidies. However, despite these efforts, there has been limited success in altering eating behaviours, particularly among categories of vulnerable citizens. Therefore, it is crucial to explore alternative strategies to address this challenge. This study is part of the OnFoods project (www.onfoods.it, National Recovery and Resilience Plan, PNRR), and focuses on designing and testing a nudging strategy tailored to encourage healthier and more sustainable dietary choices among vulnerable populations.

Method:

The different dimensions of the healthy eating nudges (cognitive, affectively and behaviourally-oriented) imply an initial choice of the strategy to be adopted. The target group of the present study consists of a subgroup of Italian fragile consumers, i.e. low socio-economic status (SES) individuals. Through initial exploratory interviews with stakeholders from diverse Charitable Associations offering food-related services, we aim to analyse service dynamics, user interactions, and cultural characteristics of ethnic groups frequenting these environments. This analysis will serve as the foundation for crafting customized nudging interventions which will be tested in field experiments conducted within canteen settings or solidarity groceries, with a focus on SES individuals. The aim is to encourage the selection of healthier and more sustainable food options. The data collection will be performed in June 2024.

Results:

Data analysis from the experimental phase will yield insights into the effectiveness of different nudging strategies on the target population. Furthermore, we will identify the connections between specific interventions and individual characteristics (e.g. socio-demographics, personal values, health consciousness, sustainability awareness). In addition, we will explore how environmental factors (including purchasing and consumption contexts, time constraints), influence these relationships.

Conclusion:

The study's results will illustrate effective nudging strategies aimed at promoting sustainable food choices among Italy's vulnerable population. Additionally, they will offer insights into the factors that hinder or facilitate these interventions, thereby informing the development of policies and practical recommendations.

Mitigating Acrylamide Formation in Cereal-Based Cookies: Impact and Efficacy of Asparaginase Treatment

Msc Shpresa Musa¹, Prof Katharina Scherf^{1,2,3}

¹Karlsruhe Institute of Technology, ²Leibniz Institute for Food Systems Biology at the Technical University of Munich, ³Technical University of Munich, TUM School of Life Sciences, Professorship of Food Biopolymer Systems

Aim:

The safety of cereal-based products is compromised by a processing contaminant formed during high-temperature processing, acrylamide (AA). AA has been classified as “probably carcinogenic to humans” and its mitigation in food is essential. Our aim was to apply asparaginases in cookie dough made of five different cereals for the reduction of AA in the final product and evaluate the potential of different cereals regarding AA formation. Additionally, the impact of varying amounts, incubation times, and temperatures of asparaginases on reducing AA and the effect of baking temperature and time was assessed. Color, texture and sensory properties were also evaluated.

Method:

Four different asparaginases were evaluated using two different concentrations, respectively. The color of baked cookies was measured using two devices, C-Cell and Color Muse. The texture was evaluated using a texture analyzer and AA was quantified using an AA ELISA kit. Finally, the sensory properties were evaluated.

Results:

Our results indicate that AA forming potential is different in wheat, rye, oat, corn, and rice cookies. This was due to the different free asparagine levels in the flour. Rye, oat, and corn cookies exceeded AA benchmark levels (350 µg/kg), however, a sufficient reduction of AA was achieved with up to 97% in oat cookies. Incubation time and temperature of asparaginases had no significant effect on AA reduction, therefore the shorter time (10 min vs. 30 min) and lower incubation temperature (60 & 70 °C vs. 90 °C) were. A significant correlation between free asparagine and AA content was found, whereas AA and the color of the final product were only weakly correlated. Overall, asparaginases showed minimal effects on cookie color and texture, with no significant changes in sensory evaluation.

Conclusion:

Our study demonstrates the critical need for mitigating AA in cereal-based products due to its carcinogenic potential. Through the application of asparaginases in cookie dough made from various cereals, significant reductions in AA levels were achieved. Additionally, the evaluation of color, texture, and sensory properties confirms the feasibility of using asparaginases without compromising the overall quality of the final product.

Contact: shpresa.musa@kit.edu

This work is part of my PhD thesis.

Are we ready for the transition towards a more sustainable diet in Europe?

Phd Ana Baranda¹, PhD Noelia Da Quinta¹, Yolanda Rios¹, Patricia Rioja¹, PhD Maria E Heikkilä², Maijaliisa Erkkola², Kamilla Bargiel-Matusiewicz³, PhD, adjunct professor Jelena M Meinilä², PhD Natalia Ziolkowska³, Agnieszka Łys³, Elena Santa Cruz¹

¹Azti Food Research Division, ²Department of Food and Nutrition, University of Helsinki, ³Faculty of Psychology, University of Warsaw

Aim:

Health and environmental concerns have intensified the urgency towards a food system that not only keeps people healthy, but also ensures food accessibility and minimises environmental impacts. Within the European Union, families with children represent a large and important consumer group (65 million households). Understanding the barriers and drivers that influence a family's dietary behaviour considering geographic, cultural, and socioeconomic differences is key to providing solutions that help their transition towards more sustainable diets.

Method:

With this aim, a cross-cultural study was carried out in three countries -Spain, Poland, and Finland- each differing in geographical location, culture, and socioeconomic characteristics, with 240 families/country. A questionnaire was launched with over 40 questions exploring knowledge, behaviour, attitude, interest, and barriers related to sustainable eating.

Results:

Overall, families showed notable self-reported knowledge about sustainable eating. However, when asking about different concepts related to this topic, there are still specific behaviours that remain unrelated to these practices. For example, while avoiding unnecessary food packing is related to a sustainable practice by 70% of the population in both Spain and Finland, in Poland that percentage is only 40%. Similar results were obtained when asking about buying local products. The prioritization of vegetable proteins is not associated with sustainability in any of the three countries (51% Finland, 29% Poland and 21% Spain) nor is the reduction in the consumption of red meats (58% Finland, 41% Poland and 37% Spain). Although there is consensus towards improving sustainable food practices, not all changes are well received. Families are willing to improve their consumption of plant or fish products but are reluctant to reduce their consumption of meat and dairy products. Regarding barriers, families in Finland highlighted the cost of sustainable products, while in Poland the difficulty in creating new habits. In all countries, there is greater interest in receiving information about seasonal foods, than about other terms such as carbon or water footprint.

Conclusion:

The results of the study will help design future interventions based on awareness, education and practical strategies, with which we can collectively contribute to a better future for ourselves and the planet.

Exploring consumer perception of more sustainable foods. Blind vs. informed study.

Phd Ana Baranda¹

¹Azti

Aim:

Related to the concepts of sustainability and food waste reduction, new sources of ingredients emerge in the innovation of product development. The market provides a broad range of plant-based (PB) products for those who wish to reduce their intake of foods of animal origin. Upcycled ingredients from food processes appear a viable solution not only to reduce food waste but also to obtain products with added value. The success of both types of products depends on adequate communication with the consumer to make them aware of their potential benefits.

Method:

Through the use of self-reported questionnaires, and neurosciences tool this work investigates the impact of communication regarding origin of ingredients on the emotional responses of 75 omnivorous and flexitarian women. They carried out a sensory evaluation of three cakes differing in the ingredient's origin: animal, vegetal and upcycled (conventional cake with different information: made with upcycled ingredients) in blind and informed conditions, during the natural action of eating a food.

Results:

Results showed a wide ignorance of both terms PB and upcycling. Just 18% and only 1.4% of participants knew the definition of both concepts, respectively. Although the samples elicited equivalent perceptions of pleasantness, they evoked different emotional responses, measured at a cognitive and behavioral level. In general, participants experienced different emotions while observing, smelling, handling, or consuming samples, both consciously and unconsciously, depending on whether the task was performed under the blind or informed condition. All of this influences the product preference decision, since in the blind condition, the majority of participants prefer the conventional product (69.4%) while in the informed condition, that figure drops to 18.1%, with the PB product now being the preferred (55.5%) followed by the one made with upcycled ingredients (26.4%).

Conclusion:

The methodology used in this study has shown how communicating about the added value at a nutritional and/or environmental level of the use of this type of ingredients resulted in a different evaluation of the products, in the evoked implicit and explicit emotions and in their willingness to pay. These results have important implications for product development and Marketing strategies.

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Algal fractions for food applications: sustainable strategies to obtain functional fractions

Joana Dias¹, José A. Teixeira^{1,2}, Cristina M. R. Rocha^{1,2}

¹CEB-UM - Centre of Biological Engineering - University of Minho, ²LABELS - Associate Laboratory on Biotechnology and Bioengineering, and Electromechanical Systems

Aim:

The main purpose is to valorize underexplored algal carbohydrates, for prebiotic and texturizing food applications, in alignment with 2030 United Nations Agenda for Sustainable Development, creating novel sustainable ingredients, with ameliorated nutraceutical and organoleptic properties. In a cascading biorefinery approach, the preliminary removal of non-carbohydrate valuable fractions and the valorization of hydrocolloid-depleted industrial algal residues was further investigated, aiming at zero-waste processing. Autohydrolysis was studied as a greener extraction/depolymerization alternative.

Method:

The red macroalgae *Gelidium corneum*, *Gracilaria vermiculophylla*, and *Porphyra dioica*; the industrial *Gelidium corneum* residue from agar extraction, and the diatom *Phaeodactylum tricornutum* were used.

In a cascading approach, cold-water (for phycobiliproteins), ethanolic (96% v/v, for pigments/lipids), and alkaline extractions (0.5M NaOH, for proteins), alone and in different sequences were evaluated. For the optimization of autohydrolysis carbohydrate recovery, biomasses were processed in a 4520 Stirred Pressurized Reactor (110°C to 230°C temperature range). The obtained extracts were characterized for yield, composition (FTIR-ATR, sugar profile including oligosaccharides with prebiotic potential, molecular weight), antioxidant capacity (FRAP, ABTS, DPPH), and rheological behavior/gelling properties.

Results:

Using a sequential pretreatment of cold water, ethanol, and NaOH, was more promising for *Gelidium*, with the obtained agar presenting an increased yield and gelling strength when compared to direct agar extraction. For *Porphyra* and *Gracilaria*, a combination of cold water and ethanol was optimal with a positive influence on the rheological properties and composition. For *Phaeodactylum*, all sequential treatments hinder the texturizing ability of the polysaccharides. For all biomasses, it was observed that the hours-long conventional hydrocolloid extractions could be replaced by minute-long autohydrolysis treatments without any negative impacts. When further subjected to autohydrolysis, the residual biomass from agar extraction (from the proposed sequential extractions or industrial processing) can be a source of oligosaccharides and protein, and the fermentation potential of the cellulosic residue is enhanced due to increased saccharification efficiency.

Conclusion:

This study successfully shortened the knowledge gap surrounding the potential of carbohydrate fractions from algae. It was proven that autohydrolysis can be applied to any source for multi-product valorization, originating sustainable functional food ingredients, and that algae should be processed in a multi-extract cascade approach.

Extraction of lipids using cell-rupture techniques from microalgae biomass cultivated on food waste hydrolysate

Prof. Nasser Hamdami¹, Payam Torabi, Prof. Omidvar Farhadian, Dr. Nafiseh Soltanizadeh, Prof. Alain Le-Bail¹

¹Oniris

Aim:

One-third of the annual global food production, is wasted yearly. The composition of food waste makes it a valuable raw material for the recovery of nutrients and for the production of high-value products. Additionally, recovery of nutrients from food waste reduces the organic matter that needs to be disposed of in landfill sites. Microalgae are potential key players for tackling greenhouse gas emissions and for providing feedstock for renewable energy production. However, carbon and nutrient requirements for their cultivation are major bottlenecks adding to the overall production costs. Alternatively, food waste could be used for the cultivation of microalgae after suitable pre-treatment to solubilize organic carbon polymers. This research discusses restaurant food waste hydrolysis using green solvents, valorisation of its hydrolysate by two species of microalga (*Scenedesmus quadricauda* and *Tetraselmis suecica*), and extraction of lipids using green technologies.

Method:

The methodology involves several steps: First, hydrolysis of food waste, nutrient recovery, and quantification of composition and elements. Second, Batch cultivation, growth evaluation, biomass concentration, and analysis of composition and elements. Third, Extraction using various pre-treatments, optimization through neural network modelling, and various analytical techniques to characterize lipid properties. Finally, modelling and optimizing the lipid extraction process from biomass.

Results:

The research demonstrates the potential of utilizing food waste as a valuable resource for nutrient recovery and high-value product production. By hydrolyzing restaurant food waste using green solvents, nutrient-rich hydrolysate is generated. This hydrolysate serves as a culture medium for cultivating *Scenedesmus quadricauda* and *Tetraselmis suecica*, enabling their growth and biomass production. Furthermore, lipids are successfully extracted from the cultivated microalgae biomass using green technologies.

Conclusion:

The study underscores the feasibility of converting food waste into valuable resources such as nutrients and lipids through sustainable and clean processes. By utilizing green solvents for food waste hydrolysis and adopting microalgae cultivation techniques, it becomes possible to address environmental concerns associated with food waste while simultaneously producing renewable energy resources and value-added products. These findings highlight the potential for sustainable solutions in waste management, contributing to a more environmentally friendly and economically viable future.

Cultivation of *Scenedesmus quadricauda* and *Tetraselmis suecica* on food waste hydrolysate; Optimization, and physicochemical studies

Prof. Nasser Hamdami¹, Payam Torabi², Prof. Omidvar Farhadian², Dr. Nafiseh Soltanizadeh², Prof. Alain Le-Bail¹

¹ONIRIS - GEPEA (UMR CNRS 6144), Site de la Géraudière CS 82225, 44322 Nantes cedex 3, France,

²Department of Food Science and Technology, College of Agriculture, Isfahan University of Technology, Isfahan 84156-83111, Iran

Aim:

Microalgae cultivation using restaurant food waste hydrolysates presents a promising avenue for sustainable biomass production and waste valorization. This study investigates the feasibility of cultivating two species of microalgae, [*Scenedesmus quadricauda* and *Tetraselmis suecica*], on hydrolysates derived from restaurant food waste through acidic hydrolysis.

Method:

The hydrolysates, obtained by treating dried restaurant food waste with hydrochloric acid (HCl), are evaluated as growth media for microalgae cultivation. Various parameters, including nutrient composition, pH, and inhibitor concentrations, are analyzed to assess the suitability of the hydrolysates for supporting microalgae growth. The growth performance, biomass productivity, lipid content, and overall biochemical composition of the cultivated microalgae are characterized and compared with traditional culture media.

Results:

The results provide insights into the potential of utilizing restaurant food waste hydrolysates as cost-effective and sustainable substrates for microalgae cultivation, contributing to the development of integrated biorefinery approaches for waste management and bioenergy production.

Conclusion:

The study underscores the feasibility of converting food waste into valuable resources such as nutrients and lipids through sustainable and clean processes. By utilizing green solvents for food waste hydrolysis and adopting microalgae cultivation techniques, it becomes possible to address environmental concerns associated with food waste while simultaneously producing renewable energy resources and value-added products.

Effect of High Pressure Homogenization on the recovery and emulsifying properties of *Chlorella pyrenoidosa* proteins

Mr. Alexandros Katsimichas¹, Dr. Maria Katsouli¹, Mr. Nikolaos Spantidos¹, Prof. Maria Giannakourou¹, Prof. Petros Taoukis¹

¹Laboratory of Food Chemistry and Technology, School of Chemical Engineering, National Technical University of Athens, Greece

Aim:

Nanoemulsions stabilized by natural emulsifiers demonstrate strong potential as delivery systems for various bioactive compounds in food and nutraceuticals. With the demand for healthier and more sustainable options growing, replacing conventional emulsifiers with natural alternatives becomes imperative. Microalgal proteins, which contain all the essential amino acids and exhibit functional properties, are considered promising as emulsifiers. *Chlorella pyrenoidosa* is a photosynthetic microalga, characterized by its high protein content (40-50%), however, due to its rigid cell wall, cell disruption is required. High pressure homogenization (HPH) is a mechanical nonthermal process in which cell suspensions are forced under high pressure through a micrometric disruption chamber, resulting in extractability enhancement and protein modifications. This study explores the impact of HPH pretreatment on recovering and enhancing properties of *C. pyrenoidosa* proteins for potential emulsifier applications.

Method:

Untreated and HPH-treated (400-800 bar, 1-4 passes) *C. pyrenoidosa* suspensions (2.5% w/w, pH=13) were incubated at 40°C for 6 h, and the supernatants were collected via centrifugation. Protein concentration was determined using the Lowry assay. Then, the supernatant pH was adjusted to 3, and proteins were collected via centrifugation and freeze-drying. Protein content was assessed using Kjeldahl method, water and oil holding capacities were determined gravimetrically and surface properties were determined through air-water surface tension measurements at 25°C using Du-Nouy ring method

Results:

At four-pass treatment, a reduction up to 12.9% of the isolate protein content was observed, due to the recovery of more intracellular compounds and cellular particles compared to untreated samples while at one-pass treatment no significant differences were observed. Increase of HPH treatment pressure significantly increased the concentration of extracted protein up to 150.9% (1-pass) and 258.5% (4-passes) compared to untreated samples. One-pass treatment, significantly increased oil holding capacity up to 16.9% compared to untreated samples, attributed to the protein unfolding and hydrophobic core exposure due to the applied high pressure. Furthermore, HPH treatment notably affected surface properties, with higher pressure and passes significantly reducing surface tension.

Conclusion:

HPH treatment enhanced protein recovery from *C. pyrenoidosa* leading to a sustainable protein extraction process. Additionally, HPH led to protein structure modification, improving their functional properties in terms of emulsion preparation.

Textural and metabolic changes in fish meat during kobujime curing

Yuri Kominami, Katio Takase, Hideki Ushio

¹The University Of Tokyo

Aim:

Several methods for dehydrating fish meat to extend its shelf life have been developed globally. One such method from Japan is known as "kobujime." In this curing technique, a raw fish fillet is sandwiched between layers of dried kelp. The fish fillet undergoes mild dehydration while simultaneously absorbing taste and odor components from the kelp. Thus, kobujime curing effectively dehydrates and seasons the fish fillet in one process. This study explored the time-course textural and metabolic changes in fish fillets during kobujime curing.

Method:

Raw flounder dorsal fillets were skinned and cut into rectangles. These flounder meat blocks were either cured with dried kelp, dehydrator sheet, or left uncured. Each block was wrapped in plastic film and stored at 4°C for varying durations. Samples of the flounder meat block before curing, and those cured for 3, 6, 12, and 24 hours, were collected. Texture profile analysis (TPA) was conducted, and moisture content was measured. Additionally, the metabolomes of the flounder meat blocks before curing and after being cured with either the kelp or dehydrator sheet for 24 hours were compared.

Results:

The time-course changes in hardness and springness of TPA parameters were similar while changes in cohesiveness and resilience were specific to the kobujime cured flounder meat. The dehydration rate varied between curing with dried kelp and dehydrator sheets. The differences in catabolic profile of the cured flounder meat according to curing method were shown in results from the metabolomic analysis. The kobujime cured flounder meat contained higher levels of saccharides.

Conclusion:

The present study investigated the effects of kobujime curing on the textural and metabolomic characteristics of flounder meat. Our results indicate that the high dehydration rate and the transfer of taste compounds from kelp induce specific textural changes in flounder meat. Additionally, these factors contribute to suppressing postmortem changes in the meat.

Pleurotus spp cultivation using algae clumps accumulated in shores of mar menor lagoon as a step forward for circular economy

Rebeca Lavega¹, S. Figueredo¹, M. Pérez², F. Marín¹, C. Soler-Rivas¹

¹Instituto de Investigación en Ciencias de la Alimentación CIAL (CSIC-UAM). C/Nicolás Cabrera 9, Universidad Autónoma de Madrid, 28049 Madrid, España.

² Centro Tecnológico de Investigación del Champiñón de La Rioja (CTICH). C/ Carretera de Calahorra km 4, 26560 Autol, La Rioja

Keywords: algae, mushrooms, substrate, cultivation

Edible mushrooms consumption should be encouraged due to their interesting nutritional value and relevant biological/medicinal properties. For their commercial production, sawdust- or straw-based substrates submitted to prior sterilization or pasteurization are commonly used. The selection of raw material varies depending on the long-term regional availability. In the last years, the lack of precipitations in the Iberian Peninsula resulted in a dramatic reduction of wheat straw availability. Simultaneously, the eutrophication process suffered by Mar Menor lagoon (Murcia) by anthropogenic causes, is inducing an uncontrolled proliferation of invasive algae such as *Caulerpa prolifera*, *Posidonia oceanica* and a mixture called "ova" including even sweet water species i.e. *Ulothrix flaca* (most abundant), *Chaetomorpha linum* (rare), *Ulva prolifera* and *Cladophora vagabunda* generating an environmental and socio-economic problem for the region.

This work aims to study the potential use of *Caulerpa prolifera* as an alternative to the conventional raw material used as mushroom substrate. A preliminary crop trial has been undertaken in order to test different percentages of algae and raw materials for the cultivation of some varieties of *Pleurotus*, such as *Pleurotus ostreatus* (PO), *Pleurotus eryngii* (PE), *Pleurotus sajor-caju* (PS) and *Pleurotus citrinopileatus* (PC). The starting substrate, sterilized for 2 hours at 120°C has been characterized from the physicochemical and structural point of view and compared with the nutritional requirements of a conventional substrate.

Results indicated that all the substrates including a proportion of *C. prolifera* contain more interesting properties such as nitrogen, minerals, lignin and hemicellulose compared to the conventional substrate. These results advance the more than plausible possibility of this material as an alternative to animal-derived compounds such as chicken manure or nitrogen supplements in mushroom cultivation.

This research was financed by the Algarikon Project (TED2021-129591B-C31). These results are under the protection of the patent nº 202430026 (15/01/2024) titled "Sustrato a base de vegetales marinos apto para el cultivo de hongos comestibles".

Development of functional noodles with *Ulva australis* to control the glycemic response

MS student Seonghyeon Nam¹, Young-Ok Son², Jongbin Lim¹

¹Department of Food Bioengineering, Jeju National University, ²Department of Animal Biotechnology, Faculty of Biotechnology, Jeju National University

Aim:

The aim of this study is to upcycle harmful alga bloom-causing *Ulva australis* as a dietary fiber source into noodle-making for the control of glycemic response.

Method:

Ulva australis (1, 5, and 10% w/w flour base) was utilized as a dietary fiber source to design the functional noodles for the control of glycemic response. The additive effects of *Ulva australis* on the noodle-making were systematically investigated by analyzing pasting property of flour, protein network formation in dough system, water hydration property during cooking process, appearance and textural property of noodles, *in vitro* carbohydrate digestibility, and postprandial glycemic response in a mouse model.

Results:

The noodles containing *Ulva australis* had a higher amount of dietary fiber inducing the distinct characteristics compared to the control. The addition of *Ulva australis* caused a low level of starch gelatinization, decreasing the peak viscosity of RVA profile, and it led to a weakness in protein network formation, showing lower C2 value on Mixolab curve. When *Ulva australis* was applied to the noodle-making, it induced a change in color, getting darker noodles compared to the control due to its natural deep green color. In the texture test, the cooked noodles treated with *Ulva australis* showed the decreased extensibility with increasing levels of *Ulva australis*, having lower R_{max} and E values. It is likely results from higher water solubility of noodles during cooking process, causing the softer texture. On the *in vitro* carbohydrate digestibility, the noodles containing *Ulva australis* had the reduced digestible fraction while the increased non-digestible fraction due to the higher amount of dietary fiber. Furthermore, it was confirmed that the consumption of the noodles produced the lower postprandial glycemic response with a blunted glucose peak using a mouse model.

Conclusion:

Overall, this study suggests new insights into how to use or upcycle harmful algal bloom-causing *Ulva australis* to design carbohydrate-based functional foods for the postprandial glycemic response modulation to manage obesity and diabetes.

Aqueous extraction of proteins and other components from microalgae during High Pressure Homogenisation treatment

Phd Student Astrid Penicaut^{1,2}, Sarah ABIDH², Rohit Srivastava³, Isabelle Chambaud², Paul MENUT¹, Veronique BOSCH¹

¹UMR Sayfood (AgroParisTech, INRAE, Université Paris-Saclay), ²Olala!, ³UMR CSGA (AgroSup Dijon, CNRS, INRAE, Université Bourgogne Franche-Comté)

Aim:

With increasing interest in sustainable protein alternatives to conventional animal or plant based proteins, the food industry seeks novel sources of ingredients. Microalgae have emerged as a promising solution for nutritional, economical and sustainable protein source. The key macromolecules are found within the cell wall and inside the cells which requires cell disruption. Previous works on microalgae cells disruption and macromolecules recovery investigated on different mechanical methods among which high pressure homogenization (HPH). The present study aims to explore simple and effective process using this technique in mild conditions for extracting the cellular biomass and solubilising constituents in the aqueous phase. The target is to understand the release processes of the cellular components as a function of the processing conditions.

Method:

Spray-dried *Chlorella* were suspended in water at different concentrations between 5-30% w/w. The suspensions were first homogenized using rotor-stator to disrupt aggregates and hydrated overnight at 4°C. Then they were heated at different temperatures during 1 hour before treated at 1000 bar from 1 to 3 passes with a HPH (Panda Plus 2000). The effectiveness of the process was qualified by particle size distribution measurements and quantified by the yield of solubles in the aqueous fraction, including proteins. Rheological measurements were carried out for supernatant viscosity.

Results:

- 1) The concentration does not affect the efficiency of the process within the studied range;
- 2) The solubilization of cell components increase from 34% to 53% as the temperature of the initial suspension increases from 4°C to 80° with 1 passes through the HPH;
- 3) The increase in number of passes increases components solubilization, taking into account the temperature increase during 3 passes;
- 4) Despite the various applied conditions, the ratio of solubilized protein to the solubilized constituents remains low suggesting the solubilization of other polymers (such as polysaccharides), as confirmed by the viscosity of the supernatants;

Conclusion:

It can be concluded that there is a synergy between temperature and number of passes during this process to solubilise the protein and other constituents of microalgae regardless of the initial concentration.

Drying of microalga (*Porosira glacialis*) derived from an industry-scale bioreactor: Energy-efficiency, product quality & stability

Sophie Kendler¹, Tom Ståle Nordtvedt¹

¹Sintef Ocean As

Aim:

This study assessed different drying methods for their energy efficiency in dewatering and drying the diatom *Porosira glacialis* produced in a scale-up setting. Additionally, the study aimed to achieve high storage stability of dried microalgae to produce a highly nutritional and stable powder for food and feed applications. Another object was to evaluate the environmental and economical sustainability of various drying methods for this microalga.

Method:

Microalga harvested from an industrial-size bioreactor underwent different drying methods (among others: spray drying, tray drying). The drying methods were cross evaluated for their energy efficiency and effect on product quality. Storage stability of dried *Porosira glacialis* was assessed by storing the powder under different environmental conditions for an extended period. Product quality parameters after drying and dewatering were evaluated, including chemical and microbial stability stored at three different water content levels. Analyses included e.g. total phenolic content, antioxidant capacity, lipid oxidation and total plate counts of yeasts, moulds and bacteria.

Results:

Significant differences in antioxidant capacity were observed at different dry matter levels. However, only minor differences were noted in other investigated product quality parameters during storage, regardless of the dryness level. Promising results were obtained for the storage of microalga with higher water content, without significant limitations on chemical or microbial stability. Economical calculations showed significant cost reductions, by minimizing drying while maintaining high product quality. Different drying methods influenced some characteristics of the powder, indicating the need for further investigation of their impact on the functional properties and interactions with other compounds in different food and feed products.

Conclusion:

This study demonstrates the great chemical and microbial stability of the diatom *Porosira glacialis* during an extended storage period, even with a high remaining water content of 40%. This suggests that less drying, and consequently less energy, is needed to generate a high-value product. Thus, a semi-dry powder can be readily incorporated into food and feed products to enhance nutritional quality. In conclusion, by evaluating the energy efficiency of different drying methods, we ensured both the environmental and economic sustainability of the process, contributing to several Sustainable Development Goals (SDGs).

Impact of Food Side-Stream Based Medium on Nutrient Composition and Protein Extractability in *Auxenochlorella protothecoides*

Mr Byron Perez^{1,2}, Dr. Alcine Chan³, Vivian Han³, Nicola Weber^{1,2}, Bruce Yong¹, Dr. Iris Haberkorn¹, Prof. Shao-Quan Liu^{3,4}, Prof. Alexander Mathys^{1,2}

¹Singapore-ETH Centre, ²Sustainable Food Processing Laboratory, ETH Zurich, ³Department of Food Science & Technology, National University of Singapore, ⁴National University of Singapore (Suzhou) Research Institute

Aim:

Assess the impact of a newly developed culture medium, designed to harness food side streams via hydrolysis, as an alternative source of nutrients to produce biocompound rich microalgae. The objective of the study was to assess the growth performance of the innovative microalgae raw material and the accumulation, composition, and extractability of the microalgae nutrients in order to show the feasibility of this cultivation system that is potentially both more sustainable and cost-effective compared to the conventional glucose-based media.

Method:

This study conducted comparative growth experiments on microalgae using three different nutrient media: a conventional low nitrogen medium (BMM_{LN}), a high nitrogen conventional medium (BBM_{HN}), and a hydrolysate medium with high nitrogen developed from brewery spent grains and soy whey hydrolysis supplemented with yeast extract (N equivalent to BBM_{HN}). The growth conditions were maintained under heterotrophic settings (50 g/L glucose). Biomass productivity, along with amino acid and fatty acid profiles, were monitored. The nutrient extractability was evaluated based on cell permeabilization and soluble proteins extraction using flow cytometry and Bicinchoninic acid (BCA) analysis after ultrasonication (20 kHz, 2.5 min, A = 61 μm).

Results:

The results showed a minimal difference between the BBM_{HN} and the BMM_{LN}, with productivities of 4.77 ± 0.05 g L⁻¹ d⁻¹ and 5.17 ± 0.78 g L⁻¹ d⁻¹, respectively. In contrast, the hydrolysate medium exhibited a longer lag phase, which resulted in slightly lower biomass productivity (4.42 ± 0.24 g L⁻¹ d⁻¹). However, the hydrolysate produced the same profile and content of amino acids and lipid as in the BBM_{HN} (peaking at $40.51 \pm 5.32\%$ and $15.21 \pm 5.30\%$ respectively). The hydrolysate medium also facilitated superior protein recoverability because of greater cell permeabilization during ultrasonication compared to both conventional media tested.

Conclusion:

Replacing traditional glucose-based media with hydrolysate from food side streams not only sustained the growth of amino acid-rich microalgae, but also enhanced the efficiency of nutrient extraction processes. This approach potentially reduces the production environmental impacts and it aligns with a more sustainable food system. The findings suggest potential for hydrolysate formulations to be used to further upscaled microalgae applications, contributing significantly to the development of novel nutrient rich food raw material sources.

Rheological and Interfacial Properties of Next-Generation Plant-based foods

Mr Abraham Badjona¹, Robert Bradshaw¹, Caroline Millman¹, Martin Howarth, Bipro Dubey¹

¹Sheffield Hallam University

Aim

Increasing interest in the design and fabrication of next generation plant-based foods that have functionalities that simulate those of conventional animal-based foods such as dairy and meat products. Differences in conformational and molecular structure of plant proteins dominate their application in different food systems. These aspects have been mostly overlooked when studying plant-based proteins to mimic animal-based foods. For manufacturers of novel plant-based products it is very important to have theoretical and experimental models to identify, predict and control key parameters impacting the textural and techno-functional attributes of these products. This research examined the influence of different structural and physical properties on the interfacial and rheological properties of commercial plant proteins compared to ultrasound extracted faba bean protein isolates.

Method

Further research into plant protein gelation and interfacial behavior is necessary to support the paradigm shift towards more sustainably produced and consumed proteins. Faba bean proteins isolate was obtained following optimization using Box Behnken Design to obtain high purity protein. To compare the rheological and interfacial properties with commercial produced plant proteins, different formulations were prepared and studied in terms of structural, physicochemical, and functional properties. Chemometric was used to examine interplay of different factors influence the rheological and interfacial properties.

Results

Major differences were observed between ultrasound extracted faba beans protein isolate compared to commercially available proteins isolates in terms of dynamic rheological parameters and interfacial properties. In addition, among the commercially available protein isolates differences were more pronounced in rheological and interfacial proteins due to structural, physicochemical, and functional differences. Under steady shear, the protein suspensions showed shear thinning behavior which were fitted to power law model.

Conclusion

The high sensitivity of plant protein systems offers the possibility of generating varying food systems with different rheological and interfacial properties depending on physicochemical and structural properties. U-faba bean protein isolate exhibited major differences in terms of rheological, structural, interfacial, and physicochemical properties when compared to commercial plant protein isolates. Additionally, interaction between protein-starch modulated several rheological and physicochemical properties. These results provide valuable information for the modulation and fabrication of next generation plant-based foods.

Towards the establishment of a recombinant production process for egg proteins

Franziska Beck¹, Nicholas Böhnlein¹, Philipp Noll¹, Marius Henkel¹

¹Technical University of Munich

Aim:

Egg proteins represent a major part in human diet and are important for food applications due to their techno-functional and nutritional characteristics, *i.a.* emulsion formation and metal-binding capacity. Consumer awareness and population growth have led to an increasing demand for sustainable, animal-free proteins driving the alternative protein research. Precision fermentation may be one potential solution for the production of egg proteins. This work focuses on the establishment of various *Escherichia coli* host systems for the specific production of ovalbumin, phosvitin and ovotransferrin. The host systems were selected based on their capacity for post-translational modifications (PTMs), *i.a.* amino acids phosphorylation, which is important for phosvitin.

Method:

A pRSET plasmid was designed for the expression of ovalbumin in *E. coli* BL21 (DE3). The initial cultivation was performed at 37 °C, and the expression was induced with 2.0 mM IPTG after 1.5 hours. Two different plasmid systems were designed for phosvitin production: 1) pRSET (high copy number) for *E. coli* BL21ΔserB and 2) pRBC (low copy number) for *E. coli* B95(DE3) ΔA ΔfabR ΔserB. The purification of the proteins will be done using His-Tag chromatographic purification (N-terminal (pRSET) or C-terminal (pRBC)). The proteins will be evaluated for their AS sequence and techno-functional characteristics.

Results:

After cloning the ovalbumin gene into the plasmid and transformation into the host system, the strain was confirmed by sequencing. The growth rate reached 0.36 1/h and the cell dry weight 175 mg/L. Ovalbumin titers of 106.21 mg/L (20 h) and a specific production rate of $q = 0.24 \text{ g/g h}$ (10 h) were achieved in shake flasks. Works on phosvitin and ovotransferrin expressions are still in progress. The data generated from these proteins and host strains are compiled in a database, along with literature data.

Conclusion:

The recombinant production of ovalbumin in *E. coli* in shake flasks was successfully achieved. To enhance the low titers, a bioreactor process must be established. However, numerous challenges remain to be addressed for each individual egg protein. These include the necessity to ensure correct PTMs, to resolve solubility and secretion issues, and to identify optimal expression conditions for subsequent upscaling.

Wholemeal pasta: Impact of drying temperature on the molecular features of macro/micro components

Senior Assistant Professor Mattia Di Nunzio¹, Alberto Barbiroli¹, Francesca Saitta¹, Davide Russo¹, Dimitrios Fessas, Stefania Iametti¹, Alessandra Marti¹

¹University Of Milan

Aim:

This study applies a multidisciplinary approach to investigate at the molecular level the relationship between product structure (defined by composition and processing) and nutritional functionality, an issue only marginally addressed in literature.

Method:

Wholemeal semolina of different protein content (12% and 15%) was used to produce pasta in a pilot-scale plant. Two different drying diagrams (low and high temperature) were applied to each semolina sample. The overall protein organization and the phenolics profile were investigated as described by Borroni et al. (10.3390/foods12102047). Differential scanning calorimetry and thermogravimetric analysis were used to assess the conformational stability changes and solvation state of macromolecules as in Emide et al. (10.1016/j.foodchem.2022.134675), whereas pasting properties were assessed as in Marti et al (10.1016/j.lwt.2013.05.008).

Results:

Proteins in the two raw materials share a similar macromolecular organization, if not for a very different distribution of hydrophobic patches on the protein surface. The drying diagram had a significant impact on the aggregation state of proteins. Increasing temperatures facilitated covalent cross-linking between storage proteins and induced thermal coagulation in cytoplasmic ones. The phenolics profile was also similar between the raw materials and was essentially insensitive to changes in the drying diagram. Instead, starch gelatinization properties and water distribution among phases already appeared to be different in the two raw materials, and such thermal properties were further affected by the different drying diagrams.

Conclusion:

These results represent a first step towards a holistic view of the role of ingredients and processing in the definition of pasta sensory and nutritional properties. This study is part of PRIN 2020 project (Grant Number 2022SCYHWK) "Combined Approaches to explore the Impact of wholemeal semolina and pasta processing on METabolic RespOnses (CALIMERO)"

Legume Grass Protein Concentrate as a Novel Ingredient.

Esteban Echeverria-jaramillo¹, Lübeck Mette², Simon G Echers², Anders K Jørgensen², Tuve Mattsson², Peter S Lübeck³, Vibeke Orlien¹

¹Department of Food Science, University Of Copenhagen, ²Department of Chemistry and Bioscience, Aalborg University, ³BiomassProtein APS

Aim: Legume grass contains the protein ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBisCo), which is gaining interest due to its complete amino acid profile and high functionality. Hence, legume grass concentrates (LGC) have potential as food ingredients. However, for LGC to pass from the laboratory to the industry, and thus be readily available in food products, a comprehensive understanding of its performance compared to existing ingredients is essential. This study aimed to assess the functional properties of LGCs, benchmarked against commonly used protein ingredients in the industry.

Method: Five different LGCs, varying in processing conditions, were obtained by ultrafiltration. This gentle process was applied to conserve the native structure of the protein. The comparison involved widely used animal proteins (whey, casein, and egg white) and plant proteins (soy and pea). Their functionality was assessed by evaluating their emulsifying and foaming properties as well as their least gelling concentration, and protein solubility.

Results: The LGC powders contained around 60% (w/w d.m.) protein. Their functionality assessment revealed that LGC exhibited high water solubility (> 90%) at pH 6 to 8, aligning with whey and egg white, and surpassing soy. The LGCs' foaming capacity at pH 7 exceeded the other ingredients and notably outperformed the 'golden standard' egg white. The overrun values of the LGCs were between 129 and 147%, while egg white overrun was only 117%. Foam stability (60 min) was 53% for the best LGC, but only 28% for egg white. Moreover, LGC's gelling capacity surpassed other proteins, forming a self-sustaining gel at a concentration of 1.5% protein (w/w), whereas egg white required at least 5% (w/w) and soy 10% (w/w).

Conclusion: The functionality of LGC aligns more closely with animal proteins (whey and egg white) than with the subpar plant ingredients. This study serves as an initial exploration into the performance of legume grass protein concentrates, suggesting their potential to replace commonly used ingredients in the industry. Ongoing efforts focus on assessing the functionality of LGC within model food systems at varying pH and ionic strength levels to identify the product categories that would benefit most from its incorporation as a plant-based protein ingredient.

The Properties of Different Plant Protein Sources: Sugar Beet and Mallow Leaves

Ayça Akyüz¹, İdil Tekin¹, Zülal Aksoy¹, **Mecit Halil Öztop**², Seda Ersus¹

¹Ege University, ²Middle East Technical University

Aim:

Today, the increasing world population raises need for sustainable and alternative protein sources. Plant-based proteins offer a more sustainable solution both economically and environmentally. Vegetable leaves, in addition to being rich in protein especially RuBisCo, have also functional properties which are important in food products formulations. As a plant protein source, sugar beet (*Beta vulgaris* L.) leaves which are a by-product of sugar production has high nutritional value with high protein content. Mallow (*Malva sylvestris* L.) is another source which is widely known as a medicinal plant with high antioxidant properties. In this study, it is aimed to compare protein contents of sugar beet and mallow leaves and some functional properties to determine the effects of different plant sources for food industry.

Method:

Total dry matter, ash, protein content and amino acid composition of sugar beet (SBL) and mallow leaves (ML) were determined. SBL protein concentrate (SBL-PC) and ML protein concentrate (ML-PC) were isolated by isoelectric precipitation. In addition to chemical composition analysis, protein solubility, emulsion activity, foaming capacity, bulk density, tapped bulk density, Carr Index and Hausner Ratio analysis were performed to ML-PC and SBL-PC.

Results:

Protein contents of SBL and ML were determined 28.26 and 40.01 g/100 g dry matter, respectively. Amino acids found in the highest amounts in SBL glutamic acid (20.67 mg aa/g protein), aspartic acid (18.15 mg aa/g protein) and leucine (15.63 mg aa/g protein) while aspartic acid (37.3 mg aa/g protein), glutamic acid (32.2 mg aa/g protein) and leucine (25.9 mg aa/g protein) in ML. Protein content of SBL-PC and ML-PC by isoelectric precipitation was determined 54.92 and 60.04 g/100 g dry matter, respectively. Also, protein concentrates differ especially in terms of their functional properties. This difference is due to the extraction conditions and the presence of other macro-components (cellulose, hemicellulose, mucilage) contained in raw material.

Conclusion:

Protein concentrates from different sources with high protein content can be used for various purposes with different protein amounts and functional properties for different food formulations. They could be both very good alternative sources in vegetarian and vegan food products such as protein bars and mayonnaise.

Production and characterization of freeze-dried WPI-lysozyme heteroprotein coacervates loaded with grape seed extract

Prof. Dr. Monika Gibis¹, Waranya Wataniyakun²

¹Dept. of Food Material Science, Institute of Food Science and Biotechnology, University of Hohenheim, ²UiT the Arctic University of Norway

Aim:

The use of heteroprotein complex coacervates (HPCC) as encapsulation and delivery systems is a topic of considerable research interest. Grape seed extract (GSE) represents an interesting ingredient for encapsulation due to its numerous health benefits. The objective of this study was to examine the impact of grape seed extract (GSE) on the coacervate formation of proteins, including lysozyme (LYS) and whey protein isolate (WPI). Furthermore, the study aimed to assess the reproducibility, sensitivity of coacervation and encapsulation efficiency. The HPCC systems were subsequently freeze-dried. The powders were analysed for structural changes using ATR-FTIR to reveal changes in the secondary structure.

Method:

Conditions for the formation of HPCC were fixed at ratio 1.5:1 (WPI:LYS) and pH 7 based on previous experiment. Heteroproteins were analysed for turbidity, ζ -potential, encapsulation efficiency, particle size. HPCC systems were freeze-dried to obtain HPCC powders, which were analysed using ATR-FTIR to show changes in secondary structure of protein and morphology using scanning electron microscope (SEM). The protein content was determined.

Results:

The addition of GSE facilitates a higher yield of coacervation, as evidenced by a turbidity of over 90 % compared to systems without GSE with less than 80 %. GSE was indeed found to be essential for coacervate formation between WPI and LYS and encapsulation efficiencies of $87.4 \pm 5.0\%$ were achieved. Coacervation is an extremely sensitive process that reacts to external conditions, e.g. ambient temperature, as shown by the large variations between batches. Freeze-drying resulted in a sheet-like structure, which may not be the best form for further applications. The presence of GSE also changes both the secondary structure of the protein, as seen in the FTIR analysis, and the appearance, as seen in the SEM.

Conclusion:

The presence of GSE influenced many factors such as the degree of coacervation, increased interactions between the two proteins used, structure of protein and the final powder yield. This was deduced to be caused by the presence of polyphenolic compounds in GSE which interacted with amino acids in WPI and LYS, enhancing the coacervate formation ability and the encapsulation efficiency of 87.4 %.

The influence of high-pressure homogenization on the techno-functional properties of commercial pea protein isolate

Yadong Li¹, Quinten Masijn¹, Olivier Goemaere¹, Ilse Fraeye¹

¹Meat Technology and Science of Protein-rich Foods (MTSP), Department of Microbial and Molecular Systems, KU Leuven Ghent Campus

Aim:

Commercial pea protein isolates (cPPI) are commonly utilized in the production of various plant-based food products. However, irreversible protein denaturation during cPPI production may negatively affect its techno-functional properties, especially its gelation potential. High-pressure homogenization (HPH) is reported as a potential technique which may improve techno-functional properties. Hence, the aim of this study is to gain mechanistic insight into the effect of HPH on thermal aggregation and thermal gelation of denatured cPPI.

Method:

Initially, 8.5% w/w cPPI dispersions homogenized at 50, 100 and 200 MPa were extensively characterized with respect to aggregation by assessing protein solubility, the composition of soluble protein fraction using size-exclusion chromatography, hydrodynamic radius through dynamic light scattering (DLS) and light microscopy. Followingly, protein structure was investigated via evaluating surface hydrophobicity, total free sulfhydryl groups, and secondary structure (Fourier-transform infrared spectroscopy). Furthermore, the HPH-treated cPPI (HcPPI) dispersions underwent thermal treatment to further study protein aggregation and the molecular interactions involved. Finally, the thermal gelation properties of HcPPI dispersions were examined through rheological tests, including temperature sweep, frequency sweep, and strain sweeps.

Results:

The application of HPH showed significant alterations in several properties of the cPPI dispersions, including improved solubility and decreased hydrodynamic radius, as confirmed by DLS and light microscopy. These results all indicated that HPH resulted in a major transformation of the large insoluble protein aggregates into smaller soluble protein aggregates. Moreover, HPH induced some changes at the molecular level, including altered secondary structure, decreased total free sulfhydryl groups, and increased surface hydrophobicity. Heating caused increased aggregation of HcPPI but did not further change the surface hydrophobicity nor the content of total free sulfhydryl groups significantly. However, the rheological tests indicated that HPH enhanced thermal gelation properties of HcPPI, especially under higher pressure conditions.

Conclusion:

This study advances the understanding of how HPH influences the structure of denatured commercial pea protein and its subsequent effects on thermal gelation properties, crucial for optimizing the use of HPH in the processing of various plant protein rich foods.

Transglutaminase crosslinking of pea protein:gelling potential to mechanistic insight through the use of proteomics

Ankita Mukherjee^{1,2,3}, Jasper Van Pee^{4,5}, Prof. Florian Weiland³, Prof. Ilse Van de Voorde³, Prof. Tara Grauwet², Prof. Ilse Fraeye¹

¹Meat Technology and Science of Protein-rich Foods (MTSP), Department of Microbial and Molecular Systems, KU Leuven, ²Laboratory of Food Technology, Department of Microbial and Molecular Systems, KU Leuven, ³Laboratory of Enzyme, Fermentation and Brewing technology, Department of Microbial and Molecular Systems, KU Leuven, ⁴ILVO Flanders Research Institute for Agriculture, Fisheries and Food, Technology and Food Science Unit, ⁵Laboratory for Animal Nutrition and Animal Product Quality, Department of Animal Sciences and Aquatic Ecology, Ghent University

Aim:

Transglutaminase (TG) is an enzyme that can covalently crosslink proteins resulting in strong gel networks, in this way contributing to structure formation in food products. This study aims to provide mechanistic insight into enzymatic crosslinking of pea proteins using transglutaminase at different dosages and the impact thereof on structure formation.

Method:

Protein dispersions (10%) were incubated up to 5 hours with 0-10 U TG/g protein at 40 °C. The enzyme was then thermally inactivated (10 min at 70 °C) after which samples were lyophilized. The degree of crosslinking was determined spectrophotometrically using the o-phthaldialdehyde (OPA) assay and the data was modelled using a fractional conversion model. SDS-PAGE analysis gave insight into participation of different pea proteins in the crosslinking reaction. The rheological properties of the gel systems were determined using small amplitude oscillatory shear. Molecular level understanding of the mechanism of crosslinking was studied with proteomics. The crosslinked proteins were enriched using SDS-PAGE, digested and analysed with mass spectrometry.

Results:

The kinetically modelled data revealed an increase in rate constant of crosslinking reaction with rising enzyme dosage indicating faster reaction. SDS-PAGE results supported this, showing a faster and more pronounced disappearance of protein bands with increased enzyme levels, indicating more extensive crosslinking. In accordance to the above methods, the rheological measurements showed a faster rise and higher end level of storage modulus G' at the end of incubation with increasing enzyme level. With proteomics, the inter- and intra-protein crosslinked sites were identified. Motif analysis revealed enrichment of glutamic acid in proximity of cross-link sites. Domain analysis revealed enrichment of cross-link sites in disordered regions of the protein.

Conclusion:

This study provides in-depth insights into the mechanism of enzymatic crosslinking of pea protein with transglutaminase. The effect of varying enzyme dosage on the structure formation was investigated and it revealed that different levels of crosslinking and gel strength can be obtained by varying the enzyme dosage and incubation time. Proteomics approach revealed insight into the localization of crosslinks at a molecular level. The results can create perspectives for the design of a variety of vegan/vegetarian products with different structural properties.

Isolation of Quinoa Proteins by Milling, Alkaline and Enzymatic-assisted Fractionations: Composition, Structure and Technofunctional Properties.

Master Student Diana Valeria Rodriguez Hernandez¹, Salwa Karboune

¹Mcgill University

Aim:

Quinoa (*Chenopodium quinoa* Willd.) stands out as an eco-friendly crop because of its resilience to harsh climate conditions, grow adaptability and minimal agricultural inputs need. Quinoa proteins have attracted attention as an alternative to animal-based sources due to the high quality of their amino acid profile and their PDCAAS score of 0.89. Additionally, their technofunctional properties make them suitable ingredients for various food applications. The present study aims at investigating the isolation of quinoa proteins by milling, alkaline and enzymatic-assisted fractionations and characterizing the compositional profiles and technofunctional properties of isolated fractions.

Method:

Quinoa varieties from Quebec were submitted to a sequential milling-assisted fractionation process leading to various fractions, F1 (embryo-rich fraction-ERF<500 µm), F2 (perisperm-rich fraction-PRF>500 µm); F3 (PRF >250µm), F4 (PRF>177µm) and F5 (PRF<177µm). For alkaline-assisted fractionation, two alkaline (2N NaOH) extractions were performed (pH9, pH11), followed by isoelectric precipitation to recover protein isolates. Enzymatic-assisted fractionation involves the use multi-enzymatic products, expressing glycosyl-hydrolases for degrading all networks surrounding the proteins. Compositional profiles and technofunctional properties of protein fractions (e.g. concentrates, isolates) were characterized and compared to that of quinoa flour.

Results:

Isolation efficiency and compositional profile of protein concentrates (PC) and isolates (PI) were dependent on the type of fractionation methods and their operating conditions. The sequential-milling-assisted fractionation process led to a PC with a protein content of 28.4% (w/w dm) in ERF- F1 fraction. Alkaline extraction resulted in PI with a protein content of 90.07%(w/w dm) at pH9 and 86.15%(w/w dm) at pH11. The protein profile of PC fractions (ERF-F1 fraction) showed lower content of 7S globulins, while that of PI (alkaline extraction at pH9) was characterized by higher content of albumins and globulins. Quinoa flour exhibited high emulsifying, swelling and water holding capacity. The fractionation process may have altered the protein structure by exposing the hydrophobic residues potentially allowing the modulation of the technofunctional properties of protein fractions.

Conclusion:

The present results contribute to the understanding of the effects of various isolation methods on quinoa proteins composition and structure. Such understanding can broaden the applications of quinoa proteins.

Analysis of Techno-functionality of Pea Protein Isolates - Understanding Processing Effects

Laura Scheuer¹, Paul Steinhardt¹, Dr. Stephanie Bader-Mittermaier¹, Prof. Dr. Ute Schweiggert-Weisz^{1,2}, Dr. Susanne Gola¹

¹Fraunhofer Institute for Process Engineering and Packaging IVV, ²Technical University of Munich, School of Life Sciences, Plant Proteins and Nutrition

Aim:

With respect to the emergence of food security issues, such as climate change, the necessity for food products to align with the concept of one-health, is rapidly growing. Especially legumes, such as pea, are well-received, owing to their positive influence on soil health along with a favorable nutritional profile. Given their increasing use in food products, it is crucial to understand and utilize the technological and functional characteristics of those protein-rich food ingredients. In the frame of the NewFoodSystems Innovation Space, we conducted a comparative study on five different pea protein isolates to elucidate discrepancies in techno-functionality. The pea protein isolates were characterized regarding their hydration and gelation properties.

Method:

Protein functionality, i.e. the ability of the protein to interact with other food product components, was evaluated by analyzing hydration and gelation properties and by correlating them with molecular characteristics. Specifically, the water hydration capacity, solubility, denaturation behaviour and particle size of protein isolate suspensions were examined. A particular emphasis was placed on the gelation process using oscillatory rheometry and the properties of the obtained gels, by assessing the least gelation concentration and performing a texture profile analysis.

Results:

Protein denaturation differences were observed via differential scanning calorimetry, possibly due to differing thermal load during processing. While water hydration capacities corresponded to expected literature values, protein solubility varied significantly among products. The least gelation concentration varied from 14 to 18% indicating differences in gel formation and stabilisation, which was corroborated by the rheological measurements. Thereby both product-specific and concentration-dependent variations occurred. Those differences could not be related to chemical composition or molecular weight distributions as those only showed slight differences for the different pea protein isolates.

Conclusion:

The selection of protein ingredients depends heavily on the intended food applications. Techno-functional characteristics, such as water hydration capacity, solubility and denaturation behavior allowed insights on the impact of varying physico-chemical properties on product performance. Based on potentially different processing techniques and techno-functional profiles of the pea protein isolates, we aim to provide valuable guidance for food manufacturers in selecting appropriate protein ingredients for their respective applications.

Uncracked byproduct valorization as multifunctional ingredients in meat substitutes

Marion Schwartz¹, Sarah Noizet¹, Nathalie Fayolle², Delphine Huc-Mathis¹, David Blumenthal¹, Marine Masson¹

¹Université Paris-Saclay, INRAE, AgroParisTech, UMR SayFood, ²JRS

Aim: Food industrial products are commonly composed of standardized pure fractions extracted and purified from raw materials, each tailored for a desired property. While this fractionation process ensures control over ingredient composition, it necessitates the use of additional solvents and dedicated purification steps, resulting in significant waste generation and potential loss of functionality. Moreover, there is growing concern about the nutritional and environmental impacts of ingredient refinement. With an increasing consumer preference for plant-based alternatives, meat substitutes are emerging. However, they often contain transformed ingredients such as coloring compounds, protein isolates and other texturing agents. To answer these challenges, we propose a novel approach to formulate meat substitutes vegetal steaks using uncracked byproducts. Our study focuses on the identification of parameters influencing both structural properties and consumer appreciation.

Methods: Apple pomace powder (APP) was used as functional ingredient (texture, color) and for its nutritional content (55% wt fibers). An experimental design was performed with 3 mix parameters (15% of the formula): water, rapeseed oil and APP and a constant fraction (85%) made from sunflower seed, onions, zucchinis, spices, red beans and quinoa. Instrumental characterization was realized by texture, color and water loss analyzes, as well as a hedonic test (24 participants) for the extreme and central recipes. Commercial references were included in these characterizations.

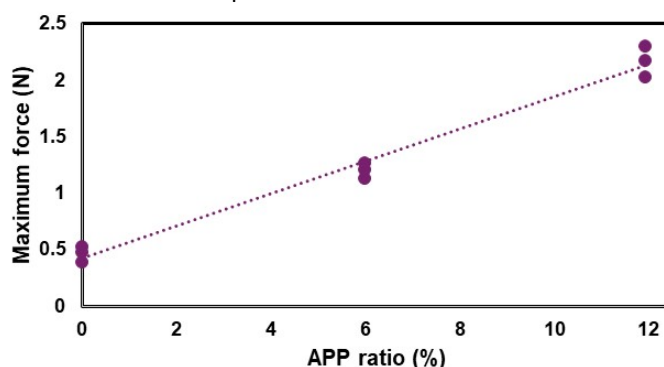


Figure 1. Maximum force (N) required to penetrate in steaks for different ratios of APP

Results: A large range of texture was obtained with only 15% of variable ingredients. The texturometry data showed that the maximum force is linearly dependant on the APP content (Figure 1). APP act as a texturizing agent, allowing to get closer to commercial steaks containing protein isolates (wheat gluten or rehydrated soy proteins). The hedonic test revealed that recipes with APP are globally appreciated. One of the formulated product has a better note than a commercial reference (note = 6.2/10)

Conclusion: This preliminary study showed very promising results and allowed the identification of improvement ways (liquid and APP ratios), paving the way to the use of uncracked byproducts in meat substitutes.

Characteristics of gels with gluten and alternative proteins to be used in meat analogues development

Phd Monica Toldra¹, PhD Dolors Pares¹, Mrs. Olga Sempere¹, PhD Elena Fulladosa²

¹Institute of Agrifood Technology, University of Girona, ²IRTA, Food Quality and Technology Program

Aim:

The main objective of this work was to carry out the physicochemical and techno-functional characterization of several sources of alternative protein to be used in high-moisture extrusion for obtaining meat analogue products.

Methods:

The products characterised were four vegetal powders: Fava Bean protein concentrate (FBC); Wheat gluten isolate (WGI), Lupine (LF), Barley (BF) and Buckwheat (BWF) flours; and one animal product: mealworm flour (MF). Solubility, oil and water absorption capacities (OAC, WAC), and the characteristics of heat-induced gels (WHC and texture) were assessed. Gels from 40% (w/v) mixtures (1:1) of WGI and the other products were prepared and compared to WGI gels.

Results:

Regarding the functional properties, the protein solubility in distilled water of the analysed products ranged from 17% (WGI and MF) to 55-60% (FBC and BWF). Solubilities at acidic pH were lower than those at neutral pH for all samples except for BF. Overall, samples showed more OAC than WAC. LF had a significantly higher hydration capacity than the other products, while the highest OAC values corresponded to WGI, BF and FBC.

The water holding capacity (WHC) of gels made of WGI suspensions (20-50% w/v) increased proportionally to the gluten content from 70% to near 100%. In terms of texture: hardness and chewiness also increased proportionally to the gluten content; nevertheless, the gluten concentration did not affect the resilience and elasticity values.

While all the gels coming from mixtures of gluten and plant flours kept WHC values higher than 98%, the insect flour led to a significant drop in this property. Concerning the texture, BWF and FBC increased the hardness and chewiness of the gels. BWF and BF increased the adhesiveness. All products, except the BWF flour, produced a significant decrease in resilience, but only the LF and MF reduced the elasticity.

Conclusion:

Addition of FBC and BWF improve the texture properties of gluten gels. Among all the mixtures evaluated, the insect flour (MF) was the product that led to gels with the lowest values of the textural attributes. Results of this work can be useful in defining the appropriate formulation to develop high moisture meat analogues.

Enhancing the nutritional and technological properties of lupine as an alternative protein by extrusion processing.

Project Manager Alba Gozalbes Sanz¹, Blanca Viadel Crespo¹, Mariana Valverde Belda¹, PhD Natalia Aparicio García¹, Elisa Gallego Vendrell¹, **PhD Lidia Tomás Cobos¹**

¹AINIA

Aim: This study aimed to investigate the potential of lupine as an alternative source of protein in response to the growing demand for sustainable and nutritious plant-based foods. The main objective of this study was to assess the impact of extrusion food processing on the nutritional value and technological and physicochemical properties of this raw material, in order to contribute to the development of healthier and more sustainable foods.

Method: The followed methodology was divided into different steps designed to achieve the proposed objectives. First, formulations containing lupine protein concentrate in combination with other pulse protein sources of interest are proposed. These were selected according to their nutritional and functional compatibility. In a second step, the extrusion process was performed on a pilot scale corotating twin-screw extruder by adjusting variables such as temperature profile, screw rotation speed, feed flow and water flow rates. These parameters were adjusted according to the behavior of the formula in the extrusion phase, in order to optimize the quality and efficiency of the process. Then, different physicochemical analyses are carried out to characterize the properties of the resulting texturized product. In the last step, the textured product was subjected a digestion process in a dynamic gastrointestinal digester, which mimics the upper gastrointestinal tract of an adult human. All the accumulated intestinal digest was subjected to a centrifugation process to obtain the soluble fraction. In the soluble fraction, the amino acid profile was analyzed to determine the digestibility of the proteins.

Results: The obtained results indicated that the extrusion process enabled the modification of the technological properties and protein digestibility of lupine. The improvement of these properties depends on the applied process conditions. In addition, an enhancement of the nutritional profile of the original raw material was observed.

Conclusion: This study represents an important step towards the valorization of lupine as an alternative protein source to develop innovative and healthy foods. These findings support the feasibility of extrusion as an effective technique to improve the technological properties and nutritional value of this legume to develop sustainable and affordable foods for the increasing global population.

Including protein quality in LCA of novel food products

Andreas Gess¹, Sara Cutroneo¹, Tullia Tedeschi¹, Mecit Oztop¹, Özlem Özmutlu Karslioglu¹, Simon Dirr¹, Lisa Ziegeltrum¹

¹Greensurvey GmbH

Aim:

Novel food products are being developed to achieve a nutritional progression while also providing an ecological advantage to their conventional counterparts. McAuliffe et al. 2022 introduced a method to integrate protein quality in the life cycle assessment (LCA) of food products based on its DIAAS score to make the possible uptake of protein from different sources comparable. For novel food products, however, a DIAAS score is hardly calculatable, as it has to be estimated for each ingredient of a commercial food product and therefore, uncertainty is multiplied with each additional ingredient. Thus, a scheme needs to be developed to include protein quality in LCA based on the amino acid composition.

Method:

A classic LCA is conducted based on the ISO norms 14040/14044 for proteins from novel sources within the ProxiMed project, e.g. chia seeds, tomato pomace, faba bean, and duckweed. Within this project, all newly developed protein products are examined on their amino acid composition in order to assess the amino acid score. The results of the nutritional assessment of the novel food products are used to establish a method that calculates a factor that represents the nutritional quality of the protein in the functional unit of the LCA.

Results:

The method is applied exemplarily on proteins from alternative sources from the ProxiMed project and their LCA results. The presented score is further developed into a general scheme of including protein quality in a nutritional functional unit based on the amino acid composition.

Conclusion:

The presented methodology represents the first approach to transfer an established method for the inclusion of protein quality in LCA into novel food products. It helps to assess the ecological aspects of novel food products more precisely in an early stage of their development. To put these products in perspective, it is vital to take in consideration their nutritional quality and digestibility when comparing them to established food products. The here presented method will be further improved to facilitate design for the environment for future novel food developments of protein products.

Understanding interactions of alternative proteins with fibre in low pH foods: Towards informing mouthfeel perception

Dr Ben Kew¹, Mrs Florence Lilley², Mr Robin Slater², Prof Anwesha Sarkar¹

¹University Of Leeds, ²Fruit Towers, Innocent Drinks

Aim:

There is increasing interest to develop protein-rich products using plants and microalgae due to their sustainability and rise in veganism. However, alternative proteins in fibre-rich products have attracted limited attention to date. Formulating fibre-rich products with plant proteins in low pH environments (pH<4.0) also pose considerable challenges owing to limited dispersibility. Herein, we used a suite of colloidal and *invitro* mouthfeel assessment to evaluate the performance of 20+ plant proteins within a fiber-rich model food matrices, aiming to efficiently incorporate sustainable proteins in low pH food applications.

Method:

Commercially available plant protein concentrates and isolates including legume (3 types of pea and soy, 2 types of faba, red lentil and chickpea), cereal (rice, oat, barely rice and rapeseed), tuber (3 types of potato), oil seed (hemp, sunflower and almond), and algae proteins (spirulina and white chorella), were compared. These proteins were blended at 2.5 wt% and 5.0 wt% protein content with a model fibre solution containing pectin (0.3 wt%), inulin fiber (0.8 wt%) and adjusted to pH 3.8. The solubility and particle size of the dispersions were determined. *Invitro* mouthfeel measurements were assessed using a tribometer with glass ball-on-PDMS set up to evaluate the friction coefficient of both low and high protein dispersions.

Results:

The protein-fibre dispersions exhibited significant issues with sedimentation and large particle size owing to aggregation at the low pH environment. Boundary friction coefficients (μ_{boundary}) varied significantly between both intra and inter proteins types. Among the tested proteins, at 2.5 wt%, the most lubricating proteins were soy and pea proteins ($\mu_{\text{boundary}} = 0.09-0.13$) whilst the poorest lubricating were spirulina, barley rice and red lentil proteins ($\mu_{\text{boundary}} > 0.2$). Upon increasing protein concentration to 5.0 wt%, all proteins showed increased μ_{boundary} with the most lubricating consisting of sunflower ($\mu_{\text{boundary}} = 0.15-0.17$) with soy not withstanding the lubricity at higher concentration ($\mu_{\text{boundary}} > 0.26$).

Conclusion:

Key findings demonstrate that fibres can offer viscosity and lubricity modification to plant proteins in acidic environments but create mouthfeel issues at higher concentrations (5.0 wt%). Ongoing work is looking at altering lubrication of these protein-fibre-rich formulations employing innovative colloidal processing.

Fibrous Structure Formation in Meat Analogues: Innovative Strategies for Low-Protein Ingredients

Anni Nisov¹, Pinja Pöri¹, Heikki Aisala¹, Markus Nikinmaa¹, Emilia Nordlund¹, Nesli Sözer¹

¹VTT Technical Research Centre Of Finland

Aim:

This research aimed to enhance fibrous structure formation in meat analogues made from low-protein ingredients. These ingredients are produced using dry fractionation, an environmentally friendly option that often yields lower protein purities compared to conventional wet fractionation. This study explored strategies to overcome this challenge during high-moisture extrusion processing (HMEP).

Method:

This study focused on dry fractionated oat, pea, and rapeseed protein concentrates with protein contents of 40%, 50%, and 41% respectively. The oat protein concentrate was subjected to enzymatic cross-linking and deamidation with or without pre-heating at 95 °C. The pea protein concentrate underwent a chemical pH-shift to pH 5 and 7. Similarly, the rapeseed protein concentrate was exposed to chemical pH-shift to 4 and 6 and was also fermented with *Lactiplantibacillus plantarum* and *Weissella confusa*. Following these treatments, the protein concentrates were freeze-dried and transformed into meat analogues through HMEP. The resulting extrudates were then evaluated for their fibrous structure, tensile strength, and free thiol groups. Additionally, solubility, particle size, and free thiol groups were measured from the raw materials.

Results:

Oat and pea protein concentrates, in their unmodified states, exhibited poor ability to form fibrous structures, whereas rapeseed protein concentrate demonstrated surprisingly robust fibrousness even at a modest 41% protein content. The fibrous structure formation of oat protein concentrate went through significant enhancement when enzymatic cross-linking and deamidation were combined with pre-heating. In contrast, without the pre-heating step, fibrous structure formation remained unchanged. Pea protein concentrate displayed the capability to form thin fibres when subjected to pH shift to 7. Additionally, the structure of rapeseed protein concentrate improved with chemical pH-shifting. Interestingly, fermentation, even when combined with pH-shifting, disrupted the fibrous structure formation ability of the rapeseed proteins.

Conclusion:

This study identified strategies to enhance fibrous structure formation in meat analogues made from low-protein raw materials. Chemical pH-shift and combination of enzymatic deamidation and cross-linking with pre-heating enhanced this process while fermentation disrupted it. These findings provide valuable insights for the environmentally friendly processing of low-protein ingredients for meat analogue applications, underscoring the potential of these strategies in overcoming the challenges posed by lower protein purities.

Assessing digestibility and sensory properties of Aviron winter peas compared to traditional spring varieties

Dr Clara Talens¹, Dr Saioa Alvarez-Sabatel¹, Irene Fenga², Milagros Arnal², Marella Alinovi², Prof Pau Talens², Maria Paciulli²

¹AZTI, Food Research, Basque Research and Technology Alliance (BRTA), Derio, Spain, ²Department of Food and Drug, University of Parma, Italy, ³Instituto Universitario de Ingeniería de Alimentos – FoodUPV. Universitat Politècnica de València. Camino de Vera s/n, 46022. Valencia, Spain

Aim:

In light of the projected global population increase by 2050 and the corresponding demand for sustainable protein sources, this study evaluates the potential of the Aviron winter pea variety—traditionally used as animal feed—for human consumption. We assess the viability of this cultivar, grown in the Basque region of northern Spain, for human consumption by comparing its properties with those of spring pea varieties traditionally consumed by humans.

Method:

We processed the Aviron variety peas into a powder form and conducted comparative analyses of their chemical composition, digestibility and techno-functional properties against those of standard spring pea varieties. The chemical composition, antinutritional factors (phytic acid, trypsin inhibitors, total phenolic and tannin contents), digestibility (protein and starch digestibility), and techno-functional properties (solubility, water and oil absorption, foaming and emulsifying capacity, least gelling concentration, oscillatory rheology, and thermal analysis) of pea powder from both varieties were evaluated.

Results:

The results revealed significant differences in moisture, sugar, and fat contents, with Aviron peas exhibiting higher starch (37.0 g/100 g–30.0 g/100 g) and similar protein levels (19.6 g/100 g–20.8 g/100 g) than the spring variety did. Antinutritional factors such as phytic acid (26.3 mg/g - 15.6 mg/g), phenolics (1.8 mg GAE/g - 1.2 mg GAE/g), and tannins (0.4 mg/g - 0.3 mg/g) were also more abundant in Aviron peas, potentially affecting protein digestibility (9.3% lower), starch digestibility (8.9% lower), and solubility (52.1% lower). However, Aviron peas could be valuable ingredients for human consumption due to their functional properties, particularly in food applications requiring gelling abilities and high foam capacity [14.0% - 7.33%].

Conclusion:

The Aviron winter pea variety presents a promising alternative protein source for human consumption. Aviron peas exhibit unique properties that could make them valuable ingredients in various food applications, especially those requiring high foam and emulsion stability. Further studies can be performed to evaluate technological strategies that may be useful for reducing the antinutritional content and consumer acceptance.

Pea protein soluble polysaccharide interactions with plant albumins

Phd Student Minh Tuan Tran¹, Akihiro Nakamura², Ruifen Li¹, Milena Corredig¹

¹Department of Food Science & CiFood center, Aarhus University, ²College of Agriculture Department of Food and Life Sciences, Ibaraki University

Aim:

To improve the use of side streams from pea protein production, it is important to study the functional properties of pea soluble polysaccharides. It was hypothesized that this polysaccharide could form complexes with proteins driven by electrostatics. In this work, we aimed to investigate the binding mechanism between high- and low-methoxyl pea soluble polysaccharide (HM-PPS and LM-PPS) and pea albumins (PAs) and β -Lactoglobulin (β -lg). The results were compared to those of a high-methoxyl pectin (HMP), which is a studied model system.

Method:

The molecular weight of HMP, HM-PPS and LM-PPS were analyzed using high-performance size exclusion chromatography equipped with a multi-angle light scattering and a refractive index detector. The molecular binding was studied at 25 °C, in a 5 mM citrate buffer at pH 4 using isothermal titration calorimetry. This technique is used to measure the heat generated when titrating protein solutions to the reaction cell containing polysaccharide and obtaining reaction thermodynamic parameters, accordingly. To better understand the difference between the complexes formed between these proteins and polysaccharides, their solubility as a function of pH was also conducted.

Results:

The average molecular weight and radius of gyration of the polysaccharides was 242 kDa – 54 nm, 505 kDa – 32 nm and 357 kDa – 28 nm for HMP, HM-PPS and LM-PPS, respectively. HMP has an elongated structure with less branches and mass distributed along its backbone, while HM-/LM-PPS has larger mass distributed in a compact structure with more branches. PAs were extracted from pea protein concentrate flour and mainly contained PA1 and PA2 fractions, with traces of lipoxygenase and lectins. Enthalpies of binding ranged from -3.6 to -16.6 kcal/mol and Gibbs free energies ranging from -6 to -9.4 kcal/mol, indicating exothermic reactions. Complexes formed with HM-/LM-PPS showed higher solubility than those prepared with HMP, in both β -lg and PAs as a function of pH in the range between 3 and 8.

Conclusion:

Structure and composition of both protein and polysaccharide were found to influence the steps and strengths of complexation. It was also concluded that pea soluble polysaccharides interact with albumin proteins and could create stable complexes at low pH.

Impact of Novel Processing Methods on the Molecular Interactions of Dietary Fibre in Snacks

Mrs Alia Alwedyan^{1,2}, Hannah Harris¹, Yaroslav Khimyak², Debora Saibene³, Frederick Warren¹

¹Quadram Institute Biosciences, Norwich Research Park, NR4 7UQ, ²School of Pharmacy, University of East Anglia, Norwich research Park, NR4 7TJ, ³PepsiCo International LTD, Beaumont Park, 4 Leycroft Road, LE4 1ET

Aim:

The consumption of dietary fibre has fallen below recommended levels in recent years, in parallel with an increase in the intake of highly processed, low-fibre snack foods. This dietary shift is associated with an escalation in obesity incidence. Dietary fibre, a carbohydrate resistant to human enzyme digestion, reaches the large intestine intact, where it undergoes fermentation by the gut microbiome, resulting in the production of metabolites called short-chain fatty acids (SCFAs), which play a role in suppressing appetite. In this research, we aim to develop healthier food products using novel processing techniques with our industrial partner PepsiCo to incorporate large quantities of dietary fibre into snacks. We also focus on understanding the intermolecular interactions within the food matrix, thus giving us insight into fibre solubility, water binding capacity, and digestibility.

Method:

Snacks were manufactured with two processing methods, conventional oven cooking and microwave-assisted drying. Digestion assays were carried out to quantify starch amylolysis. Samples were subjected to the INFOGEST digestion protocol to evaluate changes in composition and physicochemical characteristics under gastrointestinal conditions. Solid-state nuclear magnetic resonance (ssNMR) techniques were employed to study the molecular structure of the processed food snacks, as well as changes in their structure throughout digestion.

Results:

Our findings show that different processing methods have a different impact on the microstructure, moisture distribution, solubility, and water-binding properties of snack foods. These alterations affect their digestibility and accessibility to digestive enzymes. Notably, the addition of vegetables to processed snacks enhances their resistance to digestion by the human enzymes.

Conclusion:

Through collaboration with PepsiCo, we have utilized novel processing techniques to incorporate large quantities of vegetables into snacks, increasing their dietary fibre content. Molecular and spectroscopic analysis revealed that these snacks exhibit altered solubility and water-binding properties, impacting their digestibility and interaction with digestive enzymes. The incorporation of vegetables enhances the snacks' resistance to digestion, potentially conferring benefits to gut health and overall well-being.

This work is funded by PepsiCo, Inc. Views expressed are those of the authors and do not necessarily reflect the position or policy of PepsiCo, Inc.

Evaluating the Effect of Baking and Vacuum Drying on 3D-Printed Cereal Snacks Quality and Aroma

Miss Kristina Radoš¹, Associate professor Nikolina Čukelj Mustač¹, Associate Professor Maja Benković¹, Karla Obad¹, PhD Saša Drakula¹, Assistant professor Bojana Voučko¹, Professor Dubravka Novotni¹

¹University Of Zagreb, Faculty Of Food Technology And Biotechnology

Aim: Post-processing conditions are detrimental for final quality of 3D-printed cereal-based foods. Therefore, we aimed to compare the effect of two post-processing techniques (conventional baking (CB) or vacuum-drying (VD)) at two temperatures (120 or 160°C), on the quality of 3D-printed gluten-free cereal-based snacks.

Method: The millet/sweet potato flour-rice protein-psyllium dough was extrusion-printed into 12-layer cloud-shaped forms. The end of post-processing (10-35 min) was determined by monitoring snack weight (to constant) and water activity. Snack properties were assessed by image analysis, texture analyser, colorimeter and volatile flavour compounds by GC-MS. Twelve panellists were involved in descriptive (0-10 scale) and hedonic (1-9 scale) sensory evaluations.

Results: Snacks final moisture content was <3%. Shrinkage of snacks was dominantly influenced by post-process technique, and was significantly lower after CB (25.3%) than after VD (65.2%). While firmness was similar between snacks (5.74-8.57 N), crunchiness was higher for VD snacks (42.2 Nmm) than for CB snacks (28.7 Nmm). Snacks VD120 were darker than CB120, while at 160°C it was contrariwise. The 160°C and CB snacks were redder and yellower. As redness and darkness is related to burnt colour, sensory evaluated burnt odour and roasted flavour were higher in the snacks post-processed at 160°C. Overall flavour was the most expressed in CB and 160°C snacks, as was the sensory hardness. CB160 snacks generally had the highest intensities of the following sensory parameters: brown colour (6.9±1.9), burnt odour (1.4±2.0), overall odour (5.4±1.1), roasted flavour (5.0±1.6), overall flavour (5.8±1.4), bitter taste (4.9±2.5) and bitter aftertaste (5.8±1.7). Hence, CB160 had the lowest overall liking score (4.6±1.9), followed by VD160 (5.2±2.2), VD120 (5.9±1.9) whereas CB120 was moderately liked (6.7±0.9). Volatile compounds were affected by temperature or interaction of temperature and technique. Benzaldehyde (almondy) was the most expressed volatile compound which was higher at 160°C and in CB, as was E-2-octenal (fatty). Hexanal (grassy) was reduced at higher temperatures for CB, but increased after VD, while at 160°C 2-pentilfuran (nutty) increased in CB snacks, but reduced in VB snacks.

Conclusion: Generally, post-processing at lower temperatures proved favourable for the quality of 3D-printed snacks, with conventional baking being more favourable than vacuum-drying.

Effects of pulsed electric fields on dietary-fiber content and technological properties of carrot dietary-fiber concentrates

Msc Chemical And Biotechnological Harold Antonio Pajaro Escobar¹, Robert Soliva fortuny¹, Olga Martin Belloso¹, **Prof. Pedro Elez-Martinez¹**

¹University Of Lleida

Introduction: Dietary fiber concentrates derived from fruit or vegetable by-products hold potential as fiber-rich ingredients in food formulations. However, the presence of insoluble dietary fiber (IDF) fractions can hinder their incorporation into food products.

Aim: This study explored the application of pulsed electric fields (PEF) as a method to enhance the technological properties and increase the soluble dietary fiber (SDF) content of carrot by-product dietary fiber concentrates (CDFCs).

Method: Carrot pomace was processed into CDFCs through freeze-drying, crushing, and sieving (0.3 μm), followed by PEF treatments at 7 kV/cm using varying energy input levels (0, 3.1, 3.9, 4.6 and 5.7 kJ/kg). The technological properties (water retention capacity (WRC), oil retention capacity (ORC), cation exchange capacity (CEC), water swelling capacity (WSC) and solubility) as well as the SDF and IDF content of PEF-treated and untreated CDFCs were evaluated. Furthermore, the impacts of PEF on the physicochemical properties (particle size and Z-potential) and the structure were determined using scanning electron microscope, differential scanning calorimeter and X-ray diffraction.

Results: The technological properties of the CDFCs improved and the SDF content progressively increased with increasing applied energy, reaching a maximum at 4.6 kJ/kg and decreasing for higher energy inputs. Those optimal conditions led to significant increases ($p < 0.05$) in the SDF content (96.9%), WRC (69.9%), ORC (34.1%), solubility (73.7%), CEC (98.1%) and WSC (67.85%). Concurrently, the IDF content decreased (34.5%) compared to the untreated CDFCs. Structural analysis revealed that CDFCs treated at 4.6 kJ/kg exhibited a more porous structure, smaller particle size, and greater reduction of the crystalline region. These structural changes could lead to an increase in hydrophilic groups, porosity, and greater adsorption or binding of water and oil molecules, hence explaining the observed improvement in the technological properties. Redistribution of insoluble to soluble fiber fractions likely occurred in PEF-treated CDFCs at 4.6 kJ/kg.

Conclusions: PEF treatment at 4.6 kJ/kg has been proven to be an effective method for enhancing the technological properties and increasing the SDF content of CDFCs, thereby facilitating their potential incorporation as functional ingredients to formulated foods.

Enhancing the Techno-Functionality of Orange By-Products using Pulsed Electric Field treatments

Dr. Julia Nutter^{1,2}, Dr. Robert Soliva-Fortuny^{1,2}, Dr. Olga Martín-Belloso^{1,2}, **Dr. Pedro Elez-Martínez**^{1,2}
¹Department of Food Technology, Engineering and Science, University of Lleida, ²Agrotecnio Research Center

Aim:

The orange processing industry generates annually tons of residues, mainly peels (OP) and bagasse (OB), which are valuable sources of dietary fiber (DF). Pulsed electric fields (PEF) technology stands as promising avenue to modify the structure and composition of these resources and to enhance their techno-functional properties for diverse food applications. This study aimed to evaluate the feasibility of using PEF to obtain dried OP and OB by-products with enhanced techno-functional properties and to elucidate the underlying mechanisms.

Method:

OP and OB were PEF-treated (electric field strength: 3-5 kV/cm, pulse number: 50 or 200, pulsed width: 6 μ s, frequency: 3Hz, specific energy input: 1.97-28.80 kJ/kg), freeze-dried, and milled. Untreated by-products were used as a reference. The solubility, swelling capacity (SC), water retention capacity (WRC), oil retention capacity (ORC), DF content, total polyphenol content (TPC), mean particle size, and molecular structure (FT-IR analysis) were evaluated.

Results:

PEF enhanced OP's WRC, reaching a peak of 13.28 ± 0.95 g/g at the highest intensity treatment ($Q=28.8$ kJ/kg), while OB's WRC peaked at 9.39 ± 0.59 g/g ($Q \leq 4$ kJ/kg). SC increased by 15% (OP) and 23% (OB) with PEF treatment. Conversely, OP's solubility and ORC were decreased by PEF, while remained unchanged for OB. Changes in hydration properties were attributed to changes in the DF composition, particularly the insoluble:soluble DF ratio, where lower ratios correlated with higher solubility and lower WRC values. Furthermore, the mean particle size of OP doubled that of the untreated by-product when $Q \geq 6.4$ kJ/kg. PEF also improved the TPC of orange by-products, with the highest yields obtained at $Q \approx 6$ kJ/kg. FT-IR peak analysis revealed changes in molecular interactions between polysaccharides as a result of PEF treatments, suggesting the disruption of lignocellulosic material and exposure of hydroxyl groups, thus facilitating enhanced interactions between the food matrix components.

Conclusion:

PEF represents a promising technology for enhancing the WRC and SC of orange by-products, with high-intensity treatments yielding better technological properties for OP, and milder treatments favoring OB's properties. These improvements were associated with polymer breakdown and increased molecular interactions within these matrices.

Carbohydrate Core–Shell Electrospayed Microcapsules for Enhanced Oxidative Stability of Vitamin A Palmitate

Mrs. Elnaz Z. Fallahasghari¹, Marie Højgaard Lyng¹, Emma Espholin Gudnason¹, Kristin Munkerup², Ana C. Mendes¹, Ioannis S. Chronakis¹

¹Technical University Of Denmark, ²BASF A/S

Aim:

Vitamin A is an essential micronutrient that is rapidly oxidized. The encapsulation of Vitamin A Palmitate (AP) within the carbohydrate core-shell microcapsules by coaxial electrospay and the evaluation of oxidative stability of AP was investigated.

Method:

The core-shell microcapsules consisted of a shell of octenyl succinic anhydride (OSA) modified corn starch, maltose (Hi-Cap), and a core of ethyl cellulose (E)–AP produced by coaxial electrospay. The effect of different compounds (digestion-resistant maltodextrin, soy protein hydrolysate, casein protein hydrolysate, and lecithin dispersed in ethanol added to the core) or digestion-resistant maltodextrin added to the shell of the microcapsules on the oxidative stability of AP was assessed. The oxidative stability of AP was evaluated using isothermal and non-isothermal differential scanning calorimetry (DSC), and Raman and Attenuated Total Reflectance–Fourier Transform Infrared (ATR-FTIR) spectroscopy methods.

Results:

The coaxial electrospayed core-shell microcapsules had an average diameter of 3.7 μm and uniform morphology. The area under the isothermal curve for non-encapsulated AP was nearly three-fold higher in comparison to the area of the H-EAP sample, indicating the high oxidative stability that is provided by the carbohydrate matrix, while casein hydrolysate was the most protective compound added in the core (49-fold lower). From the non-isothermal DSC measurements, it was also observed that the casein hydrolysate (in the core) contributed the highest to the oxidative stability of AP (the enthalpy was hundred times lower than the non-encapsulated AP). The DSC data are in agreement with the Raman and FTIR results.

Conclusion:

The core-shell structure minimizes the amount of AP being present at the microcapsule surface and thus protects the AP from oxidation. The usage of ethanol as a solvent for the dispersion of the core compounds increased the hydrophobicity of the hydrolyzed proteins resulting in an enhancement of their antioxidant ability. Due to hydrophobic and hydrogen bonds interactions with AP, the casein acid hydrolysate was the most effective oxidation protective compound added in the core. The addition of the resistant maltodextrin in the shell of the microcapsule acted as filler, provided effective barrier properties, and enhanced the oxidative stability of AP.

Wild Sloe Berries for Rosé Vermouth Innovative Formulations

Ms Iranzu Zalba¹, Ms Montserrat Navarro¹, Mr Iñigo Arozarena¹, **Ms. Idoya Fernandez-pan**¹

¹IS-Food Research Institute. Universidad Publica de Navarra

Aim:

Vermouth is an alcoholic beverage with a balance between bitter and sweet, created by macerating wine with different spices and botanicals. It is a highly versatile and complex product that is closely linked to the natural resources of wine-producing regions. This is the case of Navarre (Spain), a region that is renowned for its biodiversity and the quality of its rosé wines. The objective was to develop a Navarrese vermouth that reflects the identity of the region. The base wine for this proposal is a rosé wine made from Garnacha grapes, macerated with hydroalcoholic extracts of a variety of wild plants, dominating sloe, the fruit of the blackthorn tree, which is native to the forests of Navarre.

Method:

The hydroalcoholic extracts of sloes were prepared using a 25-75% solutions of wine alcohol (Análisis Vínicos S.L; Spain) and a 4:3 (w:v) ratio. The mixtures were kept under orbital shaking (100 rpm) for up to 48 h. After this time, the extracts were filtered, characterized and finally added to the wine for fortifying. The extraction processes were monitored through changes in density (OIV-MA-AS2-01), TPI (Total Polyphenol Index) and chromatic characteristics (OIV-MA-AS2-07B). After fortifying the wine with the extracts, the vermouths were aged in 3.5 L glass jars, at 12 °C for 3 weeks; finally, they were subjected to sensory (hedonic test) and physicochemical characterization.

Results:

In the extraction process, significant changes were observed for both the percentage of ethanol in the hydroalcoholic solution and the extraction time (the higher the alcohol content and the longer the time, the higher the change in the chromatic characteristics). The best conditions were set as extraction processes with 50% alcoholic solutions for 24h at 23 °C and orbital shaking at 100 rpm.

Conclusion:

The fortification of the wine with the sloe extracts did not significantly affect the chromatic characteristics of the base wine but had a strong sensory impact on the aromatic, flavour and aftertaste profile of the samples. Consumers rated the samples positively and valued the use of wild sloes berries in the formulation of the vermouth as an identifying element of the territory.

Ochratoxin A in wine: detoxification through cold atmospheric plasma technology

Margaritis Tsirikas², Dr Sofia Chanioti¹, Pantelis Natskoulis¹, Prof Vasilis Valdramidis², **Sofia Chanioti¹**

¹Institute of Technology of Agricultural Products, ELGO-DEMETER, ²Laboratory of Food Chemistry, Department of Chemistry, National and Kapodistrian University of Athens

Aim:

To prevent OTA presence in wine and grape juice, it is essential to avoid grape contamination by toxigenic fungal species during pre and postharvest. The establishment of a HACCP system might drastically reduce mycotoxins in the final products. There are no available technologies for detoxification of that kind of products. The aim of this study was to evaluate the efficiency of cold atmospheric plasma (CAP) as a technology to detoxify Ochratoxin A (OTA) from wine and to analyze wine's quality indices.

Method:

A kinetic approach was followed to allow for optimal CAP processing selection targeting to OTA detoxification. A synthetic wine was prepared by tartaric acid (3.5% w/v) and ethanol (12% v/v) as diluted in ultra-pure water, while adjusting the pH to 3.5. Appropriate amount of OTA was inoculated into the synthetic wine up to a concentration of 5ppb.

A non-thermal (cold) plasma reactor featuring a pin-to-liquid dielectric barrier discharge (DBD) setup was used. Plasma treatment of synthetic wine was conducted at various high voltage settings (12%, 25%, 40% and 50% of the total generator voltage that was 24kV) and processing durations (1-4 min). Artificial solution of OTA at 5ppb (Artificial) was prepared with equal RONS concentrations to the ones produced in CAP-treated synthetic wine treated at 40% for 3 min, for comparison purposes.

All CAP-treated wine samples were evaluated in terms of OTA quantification by applying both HPLC and ELISA methods, H₂O₂ and NO²⁻/NO³⁻ concentrations, pH, conductivity as well as quality indices (total phenolic content, antioxidant activity, color and acidity).

Results:

After CAP exposure for 3 min at 40% of the total generator voltage, a reduction of OTA was observed by approximately 55%. Significant differences in the reduction of OTA were observed between CAP and Artificial as disinfectants. The reduction of OTA in Artificial solution was almost only 5% of the initial OTA concentration. The concentration of RONS in CAP-treated wines was increased significantly by increasing the generator voltage and the plasma treatment time.

Conclusion:

CAP affected OTA concentration; therefore, this technique may be useful in the food industry, providing an alternative solution for the reduction of OTA.

Comparative study of drying techniques for dried apple slices: process optimization and quality evaluation

Dr Marianna Giannoglou¹, Dr Varvara Andreou¹, Mr Konstantinos Panagiotis Masouras¹, Dr Pantelis Natskoulis¹, **Dr George Katsaros¹**

¹Institute of Technology of Agricultural Products ELGO-DEMETER

Aim: The aim of the research was to produce and comparatively evaluate the effect of air-, vacuum- and freeze-drying on the quality of apple slices, optimizing each drying process. The ultimate goal was to utilize misshaped, scabbed, bruised and imperfect (non-commercial for table-top use) apples for producing new products of optimal quality.

Method: Apple slices cv. '*Delicious Pilafa*' were dried under air or vacuum conditions at 40-80°C or freeze-dried for processing times till water activity (a_w) reaching a value of ~ 0.25 . During each drying process, the samples were analysed in terms of moisture and a_w . The effect of drying on the water loss, was mathematically modelled and drying rate constants were calculated. For each process the optimized conditions were selected based on analyses of a_w , colour parameters (CIELab), ascorbic acid, total phenolic content (TPC), total antioxidant activity (FRSA), hydroxymethylfurfural (HMF) production and sensory evaluation.

Results: Air and vacuum drying resulted in increased dehydration rates as temperature increased. The optimum processing conditions were selected as 60°C for 4 h for air and vacuum drying, and 16 h for freeze-drying, considering the data derived by the a_w , the color and texture measurements, the sensory evaluation and the HMF analysis. In all selected products, a_w value was ~ 0.25 , assuring shelf-stability. Air drying resulted in the most significant color change compared to the rest processes, with a simultaneous significant decrease in the concentrations of ascorbic acid, TPC, and FRSA by 26.5%, 17.6%, and 67.2%, respectively, compared to Control. Temperature appeared to be the main factor significantly affecting the concentration of HMF with air and vacuum drying processes showing no significant differences. Processing at 60°C for 4 h led to the production of ~ 4.4 mg/kg HMF, instead of 19.6 mg/kg HMF at 70°C for 3 h. The freeze-dried apples were of superior quality, significantly approaching the characteristics of the fresh fruit. The texture was the only parameter altered compared to Control, however this was also positively evaluated during sensory analysis.

Conclusion: The application of vacuum and freeze drying in fruits could offer products of superior quality by utilizing cull fruits that would otherwise be discarded.

Fermentation of Dry- and Wet-Extruded Pea Protein Texturates for Flavor Development

Maurice König¹, Jochen Weiss¹, Myriam Loeffler

¹University of Hohenheim

Aim:

The consumer experience with plant-based meat substitutes can be negatively affected by distinctive green, and beany off-flavors as well as the lack of desired meat flavors. This can be counteracted by implementing microbial fermentation as a production step. Therefore, the effect of microbial fermentation on volatile aroma-active compounds in pea protein texturates was studied.

Method:

Protein texturates were produced via high-moisture (HMPT) and low-moisture (LMPT) extrusion and fermented using *Staphylococcus carnosus* and *Kluyveromyces marxianus* for 4 weeks at 15 °C. The aroma profiles in the beginning and at the end of the fermentation were decoded via headspace solid-phase microextraction-gas chromatography-mass spectrometry-olfactometry (HS-SPME-GC-MS-O). Key odor-active compounds were identified. Absolute peak areas were calculated and compared.

Results:

A total of 20 (HMPT) and 18 (LMPT) odor-active compounds were identified in the unfermented samples. Among them, 6 common off-flavor compounds: hexanal, benzaldehyde, 2-pentyl furan, nonanal, (E, E)-2,4-decadienal and nonanoic acid. The concentrations of these compounds were significantly reduced over the fermentation period in HMPT and LMPT samples. Both used strains, *S. carnosus* and *K. marxianus* were influencing the aroma profiles of both texturate samples. Besides the off-flavor-reduction, the fermentation of LMPT with *S. carnosus* yielded in 6 new aroma-active compounds, while the HMPT sample resulted in 11. The amount of newly detected compounds in both texturates fermented with *K. marxianus* was 8 in LMPT and 7 in HMPT. *K. marxianus* exhibited higher efficacy in metabolizing compounds as well as a bigger variety of compounds that were metabolized compared to *S. carnosus*. Regarding formation of new compounds, there was a greater number and variety of compounds produced in HMPT compared to LMPT as well as by *S. carnosus* compared to *K. marxianus*.

Conclusion:

Depending on the desired characteristics, a recommendation can be made for usage of *K. marxianus* in case of occurrence of strong off-flavors in the product. However, with desire to create new flavor the usage of *S. carnosus* in combination with HMPT would be advantageous. We also suggest a potential mixed application of both strains for possible combination of both prevalent metabolic characteristics.

Stability of anthocyanins in plant proteins with fructooligosaccharides: Coacervation and spray drying study

Prof. Marcin Kurek¹, Ms Havva Aktas¹, Mr Patryk Pokorski¹, Dr Jorge Custodio-Mendoza¹

¹Department of Technique and Food Development, Institute of Human Nutrition Sciences, Warsaw University of Life Sciences

Aim:

Naturally derived colorants are in demand in food and nutraceutical industries owing to consumer preference for natural additives. Anthocyanins are commonly used as antioxidants and exhibit vivid colors; however, they are unstable under environmental stress like lighting, high heat and pH changes. This work evaluates the stabilization of anthocyanins from black carrot in plant protein matrices containing pea and rice proteins by coacervation double emulsion and spray drying with fructooligosaccharides (FOS) as a novel encapsulating agent.

Method:

Anthocyanins were first isolated from black carrots by ethanol in water (70: 30 v/v). Two plant protein solutions (pea and rice proteins) were prepared at 1 % (w/v) each. FOS was dissolved in water for 1% (w/v) solution. The coacervation process was a creation of a primary water-in-oil emulsion with plant proteins as emulsifying agent followed by a double emulsion with FOS solution. The resultant mixtures were spray-dried under optimized conditions (inlet temperature 180°C, feed flow rate 10 mL/min) to obtain microcapsules. The study employed a randomized complete block design (RCBD) with three replicates.

Results:

Stability tests over a 90-day period at 40°C and 75% relative humidity revealed greater anthocyanin stability retention in encapsulated samples than in controls. 70% and 65% in pea protein microcapsules and rice protein microcapsules improved anthocyanin stability respectively. Microscopy confirmed the microcapsules are uniform spherical structures having an average diameter of 10-15 µm. The encapsulation efficiency was about 85% for pea protein and 80% for rice protein. Functional tests such as solubility and release kinetics suggested a controlled release, especially in acidic conditions resembling the gastric environment.

Conclusion:

The use of pea and rice proteins, combined with FOS, effectively enhances the stability of anthocyanins through coacervation double emulsion and spray drying. This method proves to be a promising approach for improving the viability of anthocyanins as natural colorants in food and nutraceutical applications. The study highlights the potential of plant-based proteins and prebiotic fibers in creating functional and stable food additives.

Immobilization of *sn*-2 regioselective lipase on agarose based support with different enzyme-support interactions

Immobilization Of *Sn*-2 Regioselective Lipase On Agarose Based Support With Different Enzyme-support Interactions Jaegwan Lee¹

Immobilization of *sn*-2 regioselective lipase on agarose based support with different enzyme-support interactions Inwoo Park¹, Immobilization of *sn*-2 regioselective lipase on agarose based support with different enzyme-support interactions Jihoon Kim¹, Immobilization of *sn*-2 regioselective lipase on agarose based support with different enzyme-support interactions Jaehyeon Park¹, Immobilization of *sn*-2 regioselective lipase on agarose based support with different enzyme-support interactions Juchan Lee¹, Immobilization of *sn*-2 regioselective lipase on agarose based support with different enzyme-support interactions Juno Lee², Immobilization of *sn*-2 regioselective lipase on agarose based support with different enzyme-support interactions Phan-Shick Chang^{1,2,3}

¹Department of Agricultural Biotechnology, Seoul National University, ²Research Institute of Agriculture and Life Science, Seoul National University, and Center for Agricultural Microorganism and Enzyme, ³Center for Food and Bioconvergence, Seoul National University

Aim: Selection of immobilization support for *sn*-2 regioselective lipase

Immobilization is a crucial method for improving industrial capabilities of enzyme. Throughout immobilization process, characteristics of enzyme can be affected by nature of support and enzyme-support interactions. Among characteristics of lipase, *sn*-2 regioselectivity serves exclusive role in structured lipid production. Therefore, we attempted to select the support for *sn*-2 regioselective lipase immobilization based on lipase activity and retainment of *sn*-2 regioselectivity during immobilization using different enzyme-support interactions.

Method: Lipase immobilization, activity assay and regioselectivity assay

Supports used for lipase immobilization were octyl agarose for hydrophobic interaction, Q, SP agarose for ionic interaction and CN⁺ activated agarose for covalent bonding. Immobilization was conducted under 4°C with 2 mg enzyme/g support. After immobilization, lipase activity was measured with *p*-nitrophenyl palmitate (*p*-NPB) and olive oil. Furthermore, regioselectivity was analyzed by HPLC-ELSD equipped with CHIRALPAK IA column. Reusability of immobilized lipase was assessed with 5 cycles of hydrolysis reaction. After selecting a final candidate, assessment on chain length selectivity of immobilized lipase was performed.

Results: Lipase immobilization with different enzyme-support interactions

Lipase immobilization yield was highest for octyl agarose-immobilized lipase as $88.31 \pm 6.95\%$. In *p*-NPB hydrolysis, octyl agarose-immobilized lipase showed highest activity retention of $213.89 \pm 30.07\%$. In contrast, for olive oil hydrolysis, SP agarose-immobilized lipase showed highest activity retention of $50.00 \pm 5.39\%$ and total loss of activity was observed for CN⁺ activated agarose-immobilized lipase. HPLC analysis confirmed all candidates retained *sn*-2 regioselectivity of original lipase. Since Q and SP agarose-immobilized lipase exhibited poor reusability compared to octyl agarose-immobilized lipase, lipase immobilized on octyl agarose was selected for final candidate. Assessment on chain length selectivity showed shift in substrate preference during immobilization process.

Conclusion: Immobilized lipase with *sn*-2 regioselectivity

In this study, *sn*-2 regioselective lipase was immobilized on agarose based support with different enzyme-support interactions. Based on immobilization yield, activity retention, and reusability, octyl agarose-immobilized lipase was selected as final candidate. All candidates retained *sn*-2 regioselectivity, while chain length preference shift was observed for octyl agarose-immobilized lipase. These results can lay a foundation for the industrial applications of *sn*-2 regioselective lipase.

Properties of Ohmic Baked Goods Relevant for Industrial Implementation

Bsc. Elena Marek¹, Univ.Prof. Dr.Ing. Henry Jäger¹, MSc. Kate Waldert¹

¹Boku University

Aim:

Ohmic baking is an innovative rapid baking technology that utilizes only a fraction of the energy consumption that is required by conventional baking methods, while achieving enhanced product quality in cakes and bread. Previous studies have predominantly focused on the quality assessment of gluten-free ohmic-baked products whereas the effects of the ohmic baking process on wheat based products remain incompletely understood. Therefore, this study focused on the quality analysis of ohmic baked wheat bread and cakes in comparison to conventionally baked goods, aiming to identify relevant product properties for further industrial implementation.

Method:

Physical properties (volume, color, texture) were measured and compared for conventionally and ohmically baked products. Additionally, a sensory evaluation using a 9-point hedonic scale was conducted with 70 participants to assess the consumer acceptance. Furthermore, the storage stability (up to seven days at 20 °C in hermetically sealed bags) and staling behavior was investigated by measuring moisture migration and starch retrogradation. An enzyme mixture from Backaldrin was added to compare the impact on storage stability.

Results:

While ohmically baked goods typically attain greater volume, a lighter color, and increased springiness in texture, consumers generally preferred conventionally baked goods in the sensory evaluation. On average, ohmically baked goods were rated 1.4 points lower than conventionally baked goods in overall liking, with both types being rated positively on the liking scale. However, certain groups of participants showed a preference for ohmically baked cake and brownies over their conventionally baked counterparts.

The staling behavior of ohmically and conventionally baked bread differs fundamentally. In ohmically baked goods, the crumb and crust exhibit more similar moisture content compared to conventionally baked goods, resulting in less moisture migration. Additionally, starch gelatinization during ohmic baking is reduced, potentially due to the shorter baking time of 3 minutes, leading to decreased retrogradation during storage. The addition of the enzyme mixture had a greater impact on the storage stability of conventionally than on the ohmically baked goods.

Conclusion:

The results indicate that ohmic baking presents a promising method that could effectively be implemented in the industry, offering energy savings without compromising the quality of baked goods.

Enhancing Tomato Seed Oil Extraction and Bioactive Compound Retention Using Pulsed Electric Field (PEF) Technology

Franka Markić¹, Klara Kraljić², Višnja Stulić², Sanda Pleslić³, Nadica Maltar-Strmečki¹

¹Ruđer Bošković Institute, ²University of Zagreb, Faculty of Food Technology and Biotechnology,

³University of Zagreb, Faculty of Electrical Engineering and Computing

Aim

Food industries generate large amounts of by-products or waste, that offer economic and environmental benefits, during further processing [1]. Tomatoes, a key agro-industrial crop, produce 5.4-9.0 million tons of waste annually, mainly peels and seeds, which are rich in bioactive compounds like carotenoids and polyphenols. Tomato processing generates two different fractions of waste: peels and seeds which are rich in bioactive compounds like carotenoids and polyphenols [2]. The aim of this work was to investigate the oil composition and impact of novel non-thermal technology (pulsed electric field- PEF) on the tomato seed oil extraction.

Method

The PEF assisted extraction was conducted by HVG60/1 PEF, *Impel d.o.o.* The total carotenoid content was evaluated with HPLC, *Agilent Technologies*, SAD. Fatty acids composition in the oil was analysed using the Agilent Technologies 6890N Network GC System (Santa Clara, SAD). DSC analysis was performed using 214 Polyma Differential Scanning Calorimeter (NETZSCH-Gerätebau GmbH). The antioxidant activity was measured with the Bruker Magnostech ESR5000 spectrometer.

Results

Our results demonstrate a positive impact of the PEF treatment on tomato seed oil extraction. The best yield was obtained with 6 minutes of treatment at 1.25 kV/cm. The treated samples showed higher antioxidant activity and concentration compared to the untreated ones. GC analysis revealed that the major fatty acids in tomato seed oil are linoleic (48.19%), oleic (26.50%), and palmitic (13.09%). HPLC confirmed the presence of lycopene and β -carotene. DSC showed a short induction time and low oxidative stability, while PEF treatment did not affect these properties.

Conclusion

The results of this study show that PEF, a novel non-thermal technique, enhances the extraction of oil and bioactive compounds from tomato seeds. The PEF treatment also significantly increased the antioxidant activity and content in the oil. The fatty acid and carotenoid profiles indicate potential applications of tomato seed oil in the cosmetics and food industries.

Acknowledgments

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Development of novel controlled-delivery systems by electro spraying for co-encapsulation of probiotics and prebiotics

Vasco D.F. Martins^{1,2}, Miguel A. Cerqueira², Cristina Prieto³, Zoran Evtoski³, Jose M. Lagaron³, José A. Teixeira¹, Lorenzo M. Pastrana², Pablo Fuciños²

¹Centre of Biological Engineering, University of Minho, ²International Iberian Nanotechnology Laboratory, ³Novel Materials and Nanotechnology Group, Institute of Agrochemistry and Food Technology (IATA), Spanish Council for Scientific Research (CSIC)

Aim:

Dysbiosis in the human gastrointestinal tract (GIT) can lead to adverse health effects, as it has been associated to various diseases and gut disorders. To address this, new symbiotic formulations were developed based on the co-encapsulation of the probiotic *Lactobacillus Rhamnosus GG* (LGG) and a prebiotic using pH-responsive polymers. This approach aims to provide controlled release to specific parts of the GIT, potentially enhancing their effectiveness.

Method:

LGG's growth was tested with different potential prebiotics (Fibruline[®], fructooligosaccharides, pectin and inulin) at different concentrations (1 and 2%, w/v) in MRS broth. The new delivery systems were developed using electro spraying technique with and without encapsulated LGG. These systems were optimized using different biopolymers, namely, Fibruline[®] and shellac gum, combined with hydroxypropyl methylcellulose (HPMC) or by the addition of surfactants to improve electro spraying process. Polysaccharide solutions were characterized in terms of viscosity, surface tension and conductivity. Developed systems were evaluated in terms of morphology under different operation parameters, size, production rate and yield, and viability of encapsulated bacteria.

Results:

Fibruline[®] (2%, w/v) was identified as the prebiotic that most effectively promoted LGG's growth and was selected to further develop the microencapsulation systems. From 250 trials performed, two formulations were selected: 20% (w/v) shellac gum, 15% (w/v) Fibruline and 2% (w/v) Tween 80 (SFT); and 3% (w/v) HPMC, 3% (w/v) shellac gum and 2% (w/v) Fibruline (HSF). The addition of HPMC enhanced the physical properties of the dispersions, stabilized the electro spraying jet, and improved the production yield. Regarding particles' morphology, HSF showed greater sphericity, smoother surface and a narrower size distribution. The particles obtained with SFT and HSF presented a mean diameter of 997 ± 573 nm and 676 ± 260 nm, respectively. Upon LGG encapsulation, there was no significant difference in microcapsules' morphology and size. Viability assays showed that HSF and SFT capsules contained 1.14×10^8 CFU/g and 2.58×10^8 CFU/g, with survival rates of 73 ± 3 % and 84 ± 3 %, respectively.

Conclusion:

We developed new microencapsulation systems for the incorporation and protection of symbiotic products. Moreover, these materials were combined for the first time to develop microcapsule delivery systems using electro spraying, thus representing a novelty in scientific research.

Metabolic responses of apples to abiotic stressor - pulsed electric field

Aleksandra Matys¹, Dorota Witrowa-Rajchert¹, Artur Wiktor¹

¹Warsaw University of Life Sciences

Aim:

Abiotic stressors may appear at the stage of processing food products, their packaging, and storage. Their occurrence may be manifested by discolouration of the product (e.g., browning due to tissue disruption and intensification of oxidative processes), increased respiratory activity, intensified ethylene production, loss of water, and softening tissue. Mechanical damage to a plant immediately affects its visual quality, however over the long term it causes metabolic changes, the effects of which, in the form of broadly understood changes in quality, are detected after some time. This happens due to a signal generated at the site of the injury, which is then transferred to adjacent, uninjured tissue, where it can affect metabolic activity. The most common response to injury of a plant is both increased respiratory activity and ethylene production. It has also been shown that the action of abiotic stressors on plants can increase the accumulation of phenolic compounds in their tissues, which exhibit antioxidant activity. This research aimed to evaluate the impact of pulsed electric field (PEF) treatment on the respiratory activity and antioxidant capacity of apple tissue.

Method:

Apples were treated with PEF (electric field strength: 1 kV/cm). Three different specific energy inputs were analysed (1, 3.5, and 6 kJ/kg). In addition, untreated apples were also evaluated. Right after PEF treatment, materials were subjected to respiratory activity and antioxidant capacity analyses. Measurements were performed with 1-hour intervals throughout 3 hours.

Results:

It was observed that PEF treatment caused changes in the metabolism of apple tissue. Treated samples produced lower amount of ethylene than untreated sample. Slight changes in the antioxidant capacity and respiratory activity, i.e., consuming gaseous oxygen and releasing carbon dioxide, were also observed. The acceleration of metabolic reactions could be caused by plant stress. In turn, the reduction in this activity could result from irreversible cell damage by the PEF.

Conclusion:

Reduced ethylene production and changes in respiratory activity and antioxidant capacity are some of the responses of apples to the pulsed electric field.

This project has received funding from the National Science Centre (Poland) under the PRELUDIUM grant agreement No 2022/45/N/NZ9/02859.

Effectiveness of cold plasma as a new route for producing biofertilizers from food side streams

Miss Victoria Crespo-Torbado¹, Dr. Avelino Álvarez-Ordóñez¹, Miss Raquel Bodelón¹, Dr. Montserrat González-Raurich¹, Dr. Miguel Prieto¹, Dr. Mercedes López¹, **Dr. Márcia Oliveira**¹

¹University of León

Addressing food waste management is a significant global challenge, where approximately 20% of food is wasted in the European Union. Within this amount, 24% is generated during food processing and manufacturing. The nutrient-rich composition of by-products in the food industry offers significant potential for valorisation. This contributes to the circular economy model, wherein valuable materials once deemed waste are reintegrated into the supply chain to produce new products.

Nitrogen fixation, vital for agricultural productivity, has relied on energy-intensive processes such as the Haber-Bosch and Ostwald processes, resulting in significant CO₂ emissions. The Birkeland-Eyde process, employing arc plasma, presented an alternative, though with moderate energy efficiency. Recent advances in cold plasma technology hold potential for more efficient nitrogen fixation, particularly advantageous in agricultural contexts.

In light of the above, our main objective is to assess the viability of employing cold plasma technology to convert tuber processing wastes into high-value biofertilizers. To accomplish this objective, cold plasma coupled with a catalyst reactor was used under specific operational conditions.

Our study focuses on waste streams derived from potato and beetroot processing industries. We conducted a thorough analysis, encompassing both physicochemical and biochemical aspects, to assess the effects of cold plasma treatments. In terms of physicochemical outcomes, upon applying plasma treatment to the liquid waste streams, no changes were observed for the pH, electrical conductivity (EC) and oxidation-reduction potential (ORP) in beetroot streams, while in potato waste, the pH and ORP increased after plasma treatment. Regarding biochemical results, we observed noteworthy changes in the concentration of various ions, particularly nitrites and nitrates, in both potato and beetroot liquid waste streams compared to their initial levels before treatment.

Under the conditions studied, cold plasma technology exhibited promise in upgrading liquid food industry waste. This sustainable approach offers energy-efficient nitrogen fixation, with the potential to revolutionize food waste management and promoting sustainable agriculture practices.

An experimental setup for improving Soluble Gas Stabilisation (SGS) technology in muscle food

Henrik Øvrebø¹, Sara Esmailian¹, Jørgen Lerfall¹, Martin Steinert¹, Anna Olsen¹

¹Norwegian University of Science and Technology (NTNU)

Aim: Sustainable food packaging is hindered by short product shelf life and suboptimal food packaging technologies. Soluble Gas Stabilization (SGS) is an emerging technology that could solve these issues, utilizing CO₂ gas as a preservative agent before Vacuum Packaging (VP) or Modified Atmosphere Packaging (MAP). Also, with the recent focus on minimizing the processing of food products, CO₂ technology shows promise as an unprocessed preservation method. Previous studies show promising results for SGS together with VP or MAP. However, current SGS technology requires three hours to achieve comparable bacteriostatic conditions compared to MAP, which needs 48 hours, some of which are during transport and storage. This study utilizes a multidisciplinary approach by combining mechanical engineering, product development and food science. The study focuses on increasing the processing speed and, thus, the industrial adaptability of SGS. Similar to impinging freezers, flowing CO₂ combined with high pressure could improve process speed compared to only a static pressure chamber. Previous improvements have relied on numerical simulations; this study extends these efforts through empirical experimentation to validate these simulations.

Method: A modular, cost-effective flow chamber prototype was made to test the effectiveness of the proposed flow concept and collect experimental data. The experimental setup utilizes Tri-Clamp components, piping, pneumatics, and electronics and assesses differences between traditional static SGS and our novel flow-based approach. The design parallels a simulation, i.e., a virtual prototype, to enable simulation-based enhancements. Both flow and static samples (n=3) were exposed to 2 barG of CO₂ and analyzed using a gravimetric dissolution method against control samples.

Results: The flow samples showed similar CO₂ dissolution compared to static samples. In addition, one sample demonstrated an empirical 44.5% improvement in CO₂ dissolution, while theoretical calculations predicted a 7.1% difference compared to static samples due to the temperature and pressure differences. This suggests a potential for enhancing the design of flow-based SGS chambers.

Conclusion: The comparative analysis of CO₂ dissolution in different experimental setups supports the feasibility of further developing a flowing SGS technology. This advancement could substantially improve the practical application of SGS in industrial food processing, enhancing both sustainability and efficiency.

Are drying methods in fact changing the content of non-volatile compounds in plant materials?

Msc Natalia Pachura¹, Prof Mohsen Gavahian², Prof Antoni Szumny¹

¹Wrocław University of Environmental and Life Sciences, ²Department of Food Science, National Pingtung University of Science and Technology

Aim:

Our research focusses on the clarification of the effects of different drying techniques on the quantitative changes of non-volatile, biologically active compounds found in the leaves of *Melissa officinalis* and *Petroselinum crispum*. Despite the presumed stability of these compounds, the drying process can still induce changes in their content. Our aim is to comprehensively investigate this phenomenon, paying particular attention to extractability and using validation of analytical methods to confirm our findings. By shedding light on how drying affects these compounds, our work contributes to improved methods for preserving plant materials, which has significant implications across industries.

Method:

Four different drying methods were used in the research: convective drying, freeze-drying, vacuum-microwave drying, and microwave-convective drying. Bioactive compounds were isolated with solvents of different polarity, that is, sterols, higher terpenoids, phenolic acids, polyphenols, and fatty acids. Qualitative and quantitative analyses were performed using GCMS-QP2020 and LCMS-8045 (Shimadzu, Kyoto, Japan). For the identification of individual compounds analysed by GC-MS, a comparison of mass spectra, linear retention indices (LRIs) calculated from the n-alkane mixture, was performed with those available in NIST23 and FFNSC. For the LC-MS analysis the mass spectrometer will operate using electrospray ionisation and multiple reaction monitoring (MRM) mode in positive or negative ionisation. Optimal MS operating parameters were established using the 'optimisation method' function available in Shimadzu's Real Time Analysis LabSolutions. Quantitative analysis will be performed using the calibration curve method or the addition of an internal standard.

Results:

As a result of the study, two profiles of the main non-volatile compounds present in fresh and dried leaves of *Melissa officinalis* and *Petroselinum crispum* were determined qualitatively and quantitatively, and they are: apigenin, apigenin-7-o-glucoside, apiin, arbutin, bergapten, caffeic acid, caftaric acid, etc. The analytical methods used were validated (accuracy, precision, linearity, concentration range, sensitivity etc.). In addition, statistical processing of the data showed that drying had no direct effect on the change of non-volatile compounds in plant matrices. Research is currently underway to determine the relationship between the drying process and the extractability of the compounds.

Conclusion:

The results provide information on validated analytical methods to determine quantitative and qualitative changes in non-volatile compounds, and also determine the effect of the drying process on the extraction efficiency of bioactive compounds.

Drying is the most common method of preserving plant material. To optimize, that is, to shorten the process time and maintain better product quality, drying techniques have been developed in the last few decades toward the use of microwaves, vacuum, infrared, and mixed techniques. Due to the fact that water loss (whether at elevated temperatures, low pressure, or microwaves) during drying is always associated with a loss of volatile compounds (sometimes up to several tens of percent of the initial content), research on process optimisation has been the subject of numerous scientific publications. An interesting and not fully understood issue is the behaviour of nonvolatile, thermally stable substances from the group of e.g. phytosterols, alkaloids, lignans, saponins, phenolic acids, or polyphenols. According to their physicochemical characteristics, drying (in ranges of up to 70 °C) should not cause changes in their quantitative content. There are many papers in the literature that describe significant quantitative changes for compounds belonging to the nonvolatiles group. However, none of the works cited and known to me fully validated the analytical process, nor did they check the extractability, which is the subject of the research planned in our team. The aim of this research was to investigate how the drying process affects the quantitative changes of biologically active, organic non-volatile compounds in leaves of *Melissa officinalis* and *Petroselinum crispum*.

Phenolic compounds preservation after high pressure thermal processing in by-products: red grape pomace and pepper

Miriam Sánchez Ordóñez¹, Jorge Alexandre Saraiva², Carlos Alberto Pinto², Jonathan Delgado Adámez¹, Rosario Ramírez Bernabé¹

¹Centro De Investigaciones Científicas y Tecnológicas de Extremadura, ²Aveiro University

Aim:

Vegetable and winemaking industry generate great amounts of by-products worldwide. They can be re-utilized to produce ingredients rich in bioactive compounds. High Pressure Thermal processing (HPTP) is a recent technology to food preservation. HPTP consist in a combination of high pressure and temperature over a short holding time. The rapid temperature increases during compression (adiabatic heating) and temperature decrease in the product upon decompression could help to reduce the adverse heating effects encountered in conventional thermal technologies. HPTP reduces the microbial load while maintaining the bioactive compounds. The aim of this study is to evaluate the effect of HPTP on the bioactive compounds of red by-products (red pepper and red wine pomace).

Method:

Red pepper (var. *Franchi*) and red wine pomace (RWP) (var. *Tempranillo*) were obtained by a different local companies. Pepper and RWP were crushed and added to heat-sealed bags. Different temperature (65 °C, 75 °C and 85 °C) and high hydrostatic pressure intensities (600 MPa) were applied and compared to the non-treated and thermally treated samples (65 °C, 75 °C and 85 °C). The effect after the thermal treatment and HPTP were analysed on the total phenolic compounds.

Results:

The red by-products evaluated presented a different proximate composition. RWP presented acid pH, and higher percentages of fat, and fibre than red pepper. Contrarily, moisture content and *A_w* were higher in red pepper than in RWP. In red pepper, initial phenolic compounds (control) were maintained after HPTP and after thermal treatments at 65 and 85°C, while the treatment at 75°C significantly reduced the content. RWP phenolic compounds were significantly reduced with the 65°C/600MPa and 75°C/600MPa, while all thermal treatments (65 °C, 75 °C and 85 °C) and HPTP at the most intense conditions 85°C/600MPa presented intermediate reductions.

Conclusion:

The application of HPTP at any of the conditions assayed preserved initial phenolic compounds content in red pepper. However, RWP phenolic compounds were more sensitive to the temperature. Despite this the 75% of the initial phenolic compounds content were retained after HPTP at 85°C/600MPa. Thus, this technology would be suitable for the valorization of the by-products assayed.

Effect of water activity on microwave heating of dry chilis (*Capsicum annuum*)

Maria Elena Sosa-Morales¹, Mitzi G. Flores-Sánchez¹, Marco E. Pérez-Reyes²

¹Universidad De Guanajuato, ²Washington State University

Aim:

Chili is an important product in Mexico and for exportation. It is a source of minerals and capsaicin. Because it is commercialized dried, it seems safe and stable. However, there are reports of *Salmonella* presence in dry products, which is a bacterium associated to high moisture foods. The low water activity of dry products complicates the control of bacteria in them. The heat resistance of *Salmonella* increases with the decreasing of aw in foods. In some powders, effective inactivation of *Salmonella* requires temperatures over 70°C. Thus, the objective was to evaluate the effect of water activity (aw) in the heat treatment using microwaves on dry chilis (Chile de arbol, *Capsicum annuum*) and to determine the impact on their physicochemical properties.

Method:

The adjustment of aw was carried out in hermetic cells with different over-saturated solutions (K₂CO₃, Mg(NO₃)₂, and NaCl) on a period of seven days. aw, moisture content and color with CIELab scale were determined in chili samples. Subsequently, a thermal treatment was applied using microwaves at a power of 300 W, until the target temperature of 70°C. At the end of the heating, rapid cooling with forced air was applied and the physicochemical properties of the treated chili peppers were determined.

Results:

After conditioning, chilis exhibited aw values of 0.700, 0.500 and 0.410, with moisture contents of 14.1, 9.3 and 7.1% (w.b.), respectively. Greater heating time was required as the water activity of the chiles decreased, the heating time were 160, 300 and 220 s for batches of 10 g of chilis with aw values of 0.700, 0.500 and 0.410. The treated chiles had lower both aw and moisture contents ($p < 0.05$), decreasing to aw values to 0.443, 0.378 and 0.344, and moisture contents to 6.1, 5.9 and 5.9, respectively. Color was also affected by the treatments ($p < 0.05$), with linear decreasing for all parameter L*, a* and b*.

Conclusion:

aw plays a key rol in the heating time of chilis. Microwave heating was effective to get the target temperature of 70°C to inhibit *Salmonella* in this dry product. More studies are required to avoid colour changes due to treatments.

Life Cycle Assessment of protein extraction from *Coelastrella* sp. LRF1 biomass using Ohmic Heating technology

Vitor Sousa¹, Roberto Novais¹, Teresa M. Mata³, António A. Martins⁴, Ricardo N. Pereira^{1,2}

¹University Of Minho, ²LABBELS, ³INEGI, ⁴FEUP

Aim: Extracting protein from microalgae is challenging due to restrictions in operational conditions. Ohmic heating (OH) is an emerging processing strategy seen as advantageous to the extraction of proteins. In this study, the environmental performance associated with the extraction of proteins from *Coelastrella* biomass using OH will be assessed using the Life Cycle Assessment (LCA) methodology.

Method: An attributive cradle-to-gate study was done, for a functional unit of 1 g of protein extracted from *Coelastrella*. The environmental impacts were evaluated using the ReCiPe 2016 method with an equalitarian perspective, and the calculations were done in Simapro V9.5. Different electrical field intensities (35, 133 and 217 V/cm), treatment temperatures (60, 70 and 100 °C) and heating strategies (continuous and pulsed heating) were studied.

Results: The results show that using continuous OH instead of conventional heating (CH) for 10 minutes leads to a significant reduction in the impacts in all environmental impact categories. This result confirms that the use of OH has an environmental advantage over CH for protein extraction from *Coelastrella*. Differences were also observed in both OH and CH when the treatment temperatures of 60 and 70 °C were used, with a lower environmental impact at the higher temperature. Therefore, the results indicate that applying a pulse heating strategy instead of continuous heating contributes to a concomitantly improved extraction efficiency, with better environmental performance. In this case, the best operating conditions for protein extraction are the application of an electric field of 217 V/cm, using a fast and singular heating pulse and reaching a peak temperature of around 100 °C, with a holding time of less than 1 s. As an alternative energy scenario, the use of totally renewable energy was compared to the reference treatment. The obtained results considering this scenario demonstrated an improvement in 15 of the 18 indicators evaluated.

Conclusion: The LCA study conducted made it possible to understand that the application of a thermoelectric treatment contributes to a concomitant increase in extraction yield combined with better environmental performance when compared to processing by a strict thermal method.

Effect of cold atmospheric plasma on oxidation, antioxidant capacity and volatile compounds in pork meat

Ms Yelyzaveta Oliinychenko¹, Dr Saliha Saad¹, Dr Anastasia Kyriakoudi², Dr Ioannis Mourtzinis², Professor Marc Griffiths¹, Professor Brijesh Tiwari³, **Dr Alexandros Stratakos¹**

¹School of Applied Sciences, University of the West of England, ²Department of Food Science and Technology, Aristotle University of Thessaloniki, ³Teagasc Food Research Centre

Aim: Investigation of the effects of cold atmospheric plasma (CAP) application on the oxidation, antioxidant capacity, and volatile compounds profile of pork meat.

Method: Pork samples were treated with CAP using piezoelectric direct discharge technology, with air as inducing gas, (0, 6 and 9 min). Peroxide value (PA), aldehyde and protein carbonyl content were accessed to determine oxidation levels in pork. The concentration of total phenolic compounds (TPC) and inhibition of 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 3-ethylbenzothiazoline-6-sulfonic acid (ABTS) radicals were measured to estimate the antioxidant capacity of pork. Volatile compounds were analysed using selective ion flow tube mass spectrometry. One-way ANOVA (with Tukey HSD post hoc test) was used to assess differences between average values.

Results: Pork treated with CAP for 9 min exhibited significant increases in oxidation levels: aldehyde content (17% increase), protein carbonyl content (17% increase), and PA (35% increase), compared to untreated pork. In contrast, only the aldehyde content in pork treated with CAP for 6 min showed a significant increase (up to 21%) compared to untreated samples demonstrating the time-dependent impact of CAP treatment on oxidation. Pork treated with CAP for 6 and 9 min exhibited significantly higher levels of TPC with a 15% increase, in addition to higher inhibition of DPPH and ABTS radicals with a 5% and 24% increase, respectively, compared to untreated pork. This observed increase in antioxidant activity could be due to the release of endogenous antioxidants, after CAP treatment. In-depth analysis of 50 volatile organic compounds showed that the most prevalent volatiles in CAP-treated and untreated pork were aldehydes, alcohols, ketones, and acids. The levels of ethanediol, acetaldehyde, and methanol in CAP-treated pork showed a significant increase (~40%) when compared to untreated pork. In contrast, dimethylamine, acetic acid, and pentanal levels showed a significant decrease up to 20%.

Conclusion: CAP treatment of pork increased oxidation levels and induced alterations in some volatile organic compounds, for longer treatment times. However, it also led to increased antioxidant activity. Future research should focus on consumer acceptance of CAP-treated pork and further optimise treatment parameters to ensure meat safety and prolonged shelf-life without affecting quality.

Pulsed electric fields processing accelerates post-harvest ripening of kiwi fruits: Effect on hardness and quality

Dr. Athanasios Limnaios¹, Mr. Christos Tsatsis¹, Dr. George Dimopoulos^{1,2}, **Prof. Petros Taoukis¹**

¹Laboratory of Food Science and Technology, School of Chemical Engineering, National Technical University of Athens, ²Institute of Food Technology, Department of Food Science and Technology, BOKU University

Aim:

Kiwi fruits (*Actinidia deliciosa*) are climacteric, seasonal fruits exhibiting high respiration rates which continue post-harvest and result in excessive production of ethylene that induces fruit ripening. At the beginning of the harvest season, kiwi fruits are hard and unripe. Industrially, they are left for several weeks at room temperature to ripen and become softer, before distributing them to the market. Pulsed electric field (PEF) processing is a nonthermal technology causing cell permeabilization to fruits and vegetables via electroporation, when short pulses of a high intensity electric field are applied to plant tissues. This process results in turgor loss and tissue softening, which can be advantageous when seeking a technique to make hard, unripe fruits softer and perceived as ripe. The aim of this study was the investigation of mild PEF treatment on the acceleration of post-harvest ripening of kiwi fruits.

Method:

PEF processing was applied to kiwi fruits (cultivar Hayward) produced in Northern Greece and the quality characteristics of the fruits (hardness, colour, pH, titratable acidity, total soluble solids, reducing sugar and ascorbic acid concentrations) were monitored during storage. The application of different electric field strengths (0.2-1.0 kV/cm), numbers of pulses (0-5000), and preservation temperatures (2-12 °C) were evaluated during a storage period of up to 30 d.

Results:

Results indicated significant tissue softening and ripening acceleration when PEF was applied in kiwi fruits. The application of 100 pulses at 0.35 kV/cm and 50 pulses at 0.50 kV/cm gave the optimum results in terms of ripening acceleration and preservation of fruit quality, achieving a 30 % reduction in ripening time at 12 °C, compared to untreated kiwis. Under these treatment conditions, the quality characteristics of the treated and untreated kiwis remained indistinguishable during storage. At more intense treatment conditions, kiwi fruits became much softer, and their quality deteriorated at a much higher rate.

Conclusion:

PEF processing provides substantial acceleration of kiwi ripening, resulting in softer fruits with high quality characteristics. This technology could be effectively incorporated in an existing fruit packaging plant to shorten the period between harvesting of kiwi and other fruits of similar characteristics and market distribution.

The comparison of physicochemical and structural properties of carrots dried in laboratory and industrial scale

MSc Magdalena Trusińska¹, Aleksandra Matys¹, Katarzyna Rybak¹, Dorota Witrowa-Rajchert¹, Małgorzata Nowacka¹

¹Warsaw University of Life Sciences - SGGW, Institute of Food Sciences, Department of Food Engineering and Process Management, Nowoursynowska 159c

Aim:

During scaling up the food drying processes a lot of challenges can be faced and various discrepancies between products produced in different scales can be observed. Therefore, this study aimed at comparing carrot dried in the laboratory and industrial scale considering the physicochemical and structural properties.

Method:

Fresh carrots were washed and sliced, and then subjected to convective drying (CD, 78°C) and infrared-convective drying (IRCD, 60°C). The samples were dried with or without the pulsed electric field (PEF) pretreatment. The PEF energy was optimized in the preliminary research and equaled to 3 and 1.4 kJ/kg for CD and IRCD, respectively. Drying processes were carried out with the use of laboratory-scale dryers and a small industrial-scale mobile unit design by CEDRUS company. Afterwards, the quality of obtained samples was assessed on the basis of rehydration rate, hygroscopic properties, colour, and antioxidant activity. Additionally, the structural changes were analyzed with the use of scanning electron microtomography (SEM).

Results:

In both scales and types of drying, the PEF pretreatment made the tissue absorb less water after drying. On the other hand, rehydration rate was higher for the unpretreated samples. These differences could be caused by the structure alternations that can be observed in the SEM photos. Pairwise comparisons of laboratory-scale and industrial-scale samples showed that the total color difference was usually lower for industrial-scale drying (with the exception of IRCD without PEF) whereas the antioxidant activity, showing the preservation of bioactive compounds, was higher for laboratory-scale dried materials.

Conclusion:

To conclude, it can be claimed that the use of PEF pretreatment, as well as the method of drying, have an influence on the quality of dried carrot. Furthermore, in most cases the relationships between the samples treated in the same way in different scales have been kept.

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Sustainability assessment of the manothermosonication of liquid whole egg: comparative evaluation with conventional thermal pasteurisation

Sr. Enrique Beitia^{1,2}, Beatriz Silva^{1,3}, Sergiy Smetana^{1,4}, Vasilis Valdramidis², Kemal Aganovic^{1,4}

¹German Institute of Food Technologies (DIL e.V.), ²Department of Chemistry, National and Kapodistrian University of Athens, ³CBQF-Centro de Biotecnologia e Química Fina-Laboratório Associado, Escola Superior de Biotecnologia, Universidade Católica Portuguesa, ⁴Institute of Food Quality and Food Safety, University of Veterinary Medicine Hannover

Aim: Manothermosonication (MTS) emerges as an alternative microbial food decontamination method to traditional thermal technologies, which are known to be highly energy-demanded processes in the food industry. Previous studies of MTS in liquid whole egg (LWE) showed improved food quality when achieving microbial safety levels. However, the sustainability assessment of MTS as a preservation technology is still lacking. Therefore, this work aimed to perform an energy balance and life cycle assessment (LCA) comparison of LWE processing using traditional thermal and emerging MTS treatments as preservation strategies on industrial scale units.

Method: Energy assessment and LCA were performed considering the same level of microbial decontamination of *Salmonella* spp. and the same production capacity (100 kg/h) for continuous industrial production. Energy requirements were experimentally obtained for the MTS treatment and empirically calculated for the thermal treatment. Concerning LCA, a prospective attributional study using the "gate to gate" approach was conducted to estimate the environmental impact for a functional unit (FU) of 1 kg of pasteurised LWE packed, and ready for further sale. The SimaPro 9.5.0.2 software and IMPACT WORLD + methodology were used.

Results: Results from the energy assessment revealed that MTS processing required a 15.25% lower energy consumption (2.00 kWh/kg of LWE) and water compared to thermal processing (2.36 kWh/kg of LWE) because of the mechanisms of action of MTS, i.e., the cavitation phenomenon, avoiding pre-homogenisation and use of heating medium (water) for the pasteurisation stage. Concerning the LCA, lower scores in all the environmental impact indicators were obtained in MTS processing because of the lower electricity and water requirements. For example, the contribution to the climate change indicator, i.e., the carbon footprint of CO₂ emissions, from the LWE processing stages represented 57.3% in the MTS processing and 61.8% in the thermal processing, finding the environmental impact of the thermal pasteurisation stage to be 4.1-fold higher than in the pasteurisation by MTS.

Conclusion: Replacing traditional thermal preservation methods with emerging MTS in LWE processing is expected to reduce energy and water consumption, thus having a lower overall environmental impact and offering a more sustainable option.

Improving the decanter process during oat drink manufacture in terms of the protein yield

Jonas Körber¹, Dr. rer. nat. Adrian Körzendörfer¹

¹LAZBW

Aim:

Commercial oat drinks have relative low protein contents (0.2–0.7%) compared to cow's milk (3.3%). This may to some extent be attributed to an inefficient decanter process during the separation of the undesired solids from the oat slurry. The aim was to optimize the decanter process during the manufacture of oat-based milk substitutes in terms of the protein yield. Therefore, the impact of various process parameters on the decanter step was investigated.

Method:

An oat suspension (10% oat flour, 90% water) was prepared and separated using a pilot scale decanter centrifuge. A design of experiments with 30 trials was performed and the parameters feed, differential speed, feed temperature, weir height, and acceleration were varied to evaluate their influence on the separation process. Centrate samples were analyzed with regards to their dry matter content, protein content, particle size, protein composition (SDS-PAGE), and suspended particles.

Results:

The protein contents of the centrate samples ranged from 0.19 ± 0.02 to 1.10 ± 0.01 g/100 g. Data from the chemical analyses were used to calculate the optimal decanter parameters by response surface methodology. An acceleration of 4,000 g , a feed of 20 kg/h, a temperature of 20 °C, an inner weir diameter of 68 mm, and a differential speed of 180 min^{-1} resulted in the highest theoretical protein yield. A verification experiment was conducted with the optimized parameters. This led to the highest protein yield in view of all samples that were sufficiently clarified, i.e., free of dark particles (husk and embryo residues). SDS-PAGE further proved that none of the oat protein fractions was completely separated during the optimized decanter process.

Conclusion:

This work shows that unfavorable process parameters lead to a loss of protein during the decanter step. Hence, the parameters of the decanter process during the manufacture of oat-based milk substitutes should be set carefully in order to achieve a high protein yield. This enables both a higher process efficiency and products with improved nutritional properties.

Key physicochemical parameters of NO_x production in plasma-processed-air (PPA) for processing herbs & spices

Yijiao Yao^{1,2}, Thomas Weihe², Jörg Ehlbeck², **Dr. Uta Schnabel**, Kimon-Andreas Karatzas¹

¹University of Reading, ²Leibniz Institute for Plasma Science and Technology

Aim: Non-thermal plasma (NTP), which is recognized by its strong antimicrobial efficacy on microorganisms associated with food and food production environment, stands out as a novel and promising technology for food processing. However, the limited knowledge of the chemical composition, differences in physical and chemical processes at various upscaling levels hinders the successful transition of this technology from laboratory settings to industrial-scale applications. Therefore, this study aims to characterize the chemical composition of plasma-processed air (PPA) produced from lab-scale and pilot-scale equipment. Furthermore, the impact of different operational parameters such as humidity, power and flow rate for PPA production is to be revealed.

Method: Two in-house (INP) built microwave plasma sources (2.45 GHz), MidiPLexc and the extended version PLexc², were applied. The chemical composition of PPA was fully investigated by Fourier transform infrared spectroscopy (FTIR).

Results: Nitrogen monoxide (NO), nitrogen dioxide (NO₂) and dinitrogen pentoxide (N₂O₄) were detected in PPA, where NO₂ took up more than 70 % in quantity of the key reactive species. The NO_x (sum of NO and NO₂) concentration in PPA was 13465 ppm for MidiPLexc and 14176 ppm for PLexc². Input power and flow rate have significant impact on the production of these reactive species. A model was built for the estimation of the concentrations of the chemical species produced under different energy inputs.

Conclusion: These results provide fundamental and essential knowledge in the optimization of the control software and hardware of the plasma devices and further promote the applications of NTP in food industries.

Perception of edible insects in Hungary. Online survey among university students and graduates.

Prof. Dr. Diána Bánáti¹, Dr. Edina Lendvai¹

¹University of Szeged

Aim:

Edible insects are alternative protein sources and subject to authorisation as novel food in the European Union. There is no history of insect consumption in Europe, where neophobia, disgust and lack of information are major barriers to insect eating. The perception of edible insects, consumer attitudes through associations about insects, future options were studied among university students and fresh graduates in Hungary. We also wanted to explore the level of knowledge about edible insects and the potential ways on how to overcome aversions.

Method:

An online questionnaire was shared with the students of the University of Szeged. The main objectives of the questionnaire were to study:

- consumer attitudes through associations re edible insects,
- facts about consumption, future option,
- level of knowledge,
- future options (ways to overcome aversion, preparation methods).

440 valid responses were analysed. The research was not representative. Microsoft Excel 2016 MSO was used to evaluate the results. The options used were the frequency function, descriptive statistics and the statement chart.

Results:

The results clearly demonstrate, that consumers surveyed had very little knowledge about edible insects, but they would be open to try and taste them, in case they had more information. The first association of the respondents referred to disapproval and far-away (exotic) regions, while protein source and sustainability were also mentioned spontaneously. Interestingly, men would accept such a novel food more, than women. But no similar significant differences were observed across age groups, although young people seemed to be more receptive. In case insects would not resemble their original form, but would be presented to consumers in the form of insect flour, powder or oil, then they would be willing to accept edible insects more.

Conclusion:

The results suggest that there is a need to provide as much information as possible to the general public, highlighting the main reasons why edible insects would provide food and protein to consumers in a sustainable way.

We believe that the research we have carried out should be continued, for example in the form of a focus group study with insect tasting.

Exploring the Effects of Probiotic-Supplemented Diet on Yellow Mealworm Production: Laboratory vs. Industrial-Scale Investigation

Poster Presentation Sabina Dahal¹, Dr. Antoine Lecocq², Dr. Uri Lesmes³, Dr. Aberham Hailu Feyissa¹, Dr. Federico Casanova¹

¹Technican University Of Denmark (dtu), ²University of Copenhagen , ³Technion -Israel Institute of Technology

Aim:

Understanding the applicability of insects for feed and/or food applications is a growing and a global interest during the last years. The supplementation of beneficial bacteria in insect's diet for boosting immunocompetence and growth improvement is a new area of investigation. The objective of this study is to investigate the effect of probiotic supplementation on insect production and compare the feeding effect between laboratory scale and industrial scale.

Method:

Both laboratory and industrial scale were conducted with similar experimental setups. Similarity in indoor climate during insect rearing at both locations was maintained as close as possible with temperature of 27°C and relative humidity between 60-70%. The scale of experiment was 20 times smaller in laboratory scale compared to industrial scale which was based on beetle density used for oviposition. Three types of probiotic strains were used at three different levels of doses: recommended dose, double the recommended dose, half of the recommended dose. Probiotic-enriched feed was supplemented when larvae reached five weeks old. Larval growth was recorded weekly starting from week five until week nine when larvae were ready for harvesting. Proximate analysis as well as protein quality of the harvested larvae samples was further analyzed for all concentration doses.

Results:

Results regarding probiotic effects on larval growth, survival, total biomass, and nutritional profile will be presented and discussed at the conference.

Conclusion:

From the preliminary findings, it was found that there was slight difference in individual larval weight, but significant difference was found in harvest weight at industrial scale.

Effects of cell-free hemolymph from diapausing *Ostrinia nubilalis* larvae on human normometabolic and cancer cells

M.Sc. Teodora Knežić¹, Dr. Miloš Avramov², M.Sc. Vanja Tatić², Dr. Snežana Gošić-Dondo³, Dr. Ivana Gadjanski¹, Prof. Dr. Željko D. Popović²

¹BioSense Institute, University of Novi Sad, ²Faculty of Sciences, University of Novi Sad, ³Maize Research Institute "Zemun polje"

Aim: Insects are of great interest not only as potential solutions for the growing problem of food scarcity, but also as sources of alternative proteins and biologically active compounds for improving the quality of foodstuff. Insect larval hemolymph is rich in proteins, making it a prospective candidate for food industry-related research. In this study, we explored the potential of turning a common pest insect species, the European corn borer (*Ostrinia nubilalis*, ECB), into a beneficial one. In particular, this species is of interest because of its ecophysiological adaptations which allow it to survive long winter periods with little to no food available.

Method: Cell-free hemolymph from 5th instar diapausing larvae of the ECB was characterized in terms of antiproliferative effects on human cells. Total protein concentration of collected hemolymph was determined using the Bradford assay, after which human cancer (U-87) and normometabolic cells (MRC-5) were treated with different concentrations of hemolymph proteins (serial dilutions from 2000 µg/mL to 7.8125 µg/mL). The effects on cell viability were measured 24, 48 and 72h after treatment with the MTT assay.

Results: Results of the Bradford assay have shown that hemolymph from ECB diapausing larvae is highly rich in proteins, which is in line with the general findings on protein content of insect larval hemolymph. As for the cytotoxicity assay, it was shown that ECB larval hemolymph differently affected the viability of U-87 and MRC-5 cells in dose- and time-dependent manners. Antiproliferative effects were visible on U-87 cells already 24h after treatment with a concentration of 250 µg/mL of hemolymph proteins. Cytotoxic effects on MRC-5 were first observed only after 48h, and only after treatment with the highest concentrations of hemolymph proteins (1000 and 2000 µg/mL)

Conclusion: The results of this study show the potential anticancer effects of hemolymph from diapausing ECB larvae, as the treatments had a much stronger negative effect on the viability of the cancer cell line, in comparison to the normometabolic fibroblasts. This difference in cell type-dependent effects shows that further research on the topic of insect hemolymph characterization and applicability can be undertaken in both food- and pharmaceutical-related directions.

Sustainable Hamburger using insect-based ingredients.

Mrs. Ana Vinhas¹, Marta Coelho¹, André Mota¹, Miguel Teixeira¹

¹Colab4food

The intricate landscape of human behavior regarding dietary choices, emphasizing the subtle intersection between personal health and environmental sustainability leads people to try new foods. Insect-based foods have garnered significant attention as a viable protein alternative in recent years due to their rich nutritional profile and cost-effectiveness. This study investigates the utilization of insect flour as a novel ingredient in the development of sustainable hamburger products by reducing beef reliance in hamburgers by 30% (w/w), seeking a more sustainable product. This study is aligned with dietary guidelines advocating for moderation in red meat consumption and promoting sustainable and healthy dietary choices.

The methodology employed involved market analyses and literature reviews to identify opportunities associated to beef content reduction in hamburgers and alternative protein sources incorporation. Subsequently, formulation experiments were carried out to develop a hamburger recipe that met the study objectives while providing an appealing and unique flavor profile. Different *Tenebrio molitor* flour incorporation levels were tested (6,5% (w/w) to 16% (w/w)), to determine the optimal combination balancing sensorial and nutritional aspects. In order to keep the main objective and considering the insect flour incorporation was not proportional due to legal and regulatory limits, bulking ingredients were integrated.

The results demonstrate promising outcomes using insect flour as ingredient to provide a sustainable and flavorful substitute for traditional beef. Compared to the standard composition of a regular hamburger, the inclusion of insects entails an increase in protein content, the addition of fiber to the final product- an element absent in a typical hamburger- and a boost in mono- and polyunsaturated fatty acids.

Concluding our study, this research highlights the potential of incorporating insect flour into hamburger production. From our perspective, these findings hold particular significance within the realm of food industry and gastronomy, offering inspiration for businesses operating in the eyes of sustainability.

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Essential and non-essential elements in *Sparus aurata* fed by alternative insect and poultry-based diets

Phd Marina Cano Lamadrid¹, Miss Nuria Jiménez-Redondo¹, PhD Antonio J Signes-Pastor², PhD Paul Holhorea³, PhD Álvaro Belenguer³, PhD Jaume Pérez³, PhD Esther Sendra¹

¹Instituto de Investigación e Innovación Agroalimentaria y Agroambiental (CIAGRO-UMH), Universidad Miguel Hernández, Carretera de Beniel, km 3.2, 03312, ²Instituto de Investigación Sanitaria y Biomédica de Alicante, Universidad Miguel Hernández (ISABIAL-UMH), 03010 Alicante, Spain. Unidad de Epidemiología de la Nutrición, Departamento de Salud Pública, Historia de la Ciencia y Ginecología, Universidad Miguel Hernández (UMH), 03550 Alicante, Spain. CIBER Epidemiología y Salud Pública (CIBERESP), Instituto de Salud Carlos III, 28034, ³Nutrigenomics and Fish Growth Endocrinology Group Institute of Aquaculture Torre de la Sal. CSIC 12595

Aim: Aquaculture 2021-2030 EU strategy is focused on maintaining the nutritional quality and reducing the presence of abiotic contaminants of farmed fish, which is included in the European Green Deal and the Farm to Fork Strategy. Therefore, the aim of this research was to evaluate the effect of different formulation feed (FM based on fish meal as a control, and alternative feeds: PAP based on poultry and porcine meal, and ALT based on insect meal) used in farmed gilthead sea bream on the profile of essential (for example Ca and K) and non-essential (for example total As) elements. Additionally, based on the average human consumption in one of the main producing and consuming countries in Europe, Spain, an estimation of the intake of the essential and non-essential elements under study was carried out.

Method: Gilt-head Sea breams were fed in the IATS culture facilities from 10-15 g until reaching commercial size using FM, PAP, and ALT diet with no change in growth and feed conversion rates. After fattening, 20 fish per diet were slaughtered and sent to the CIAGRO-UMH facilities. Microwave digester was used before using ICP-MS for determination and quantification of essential and non-essential elements. For statistical analysis, an ANOVA test and Tukey's test ($p < 0.05$) were performed for comparison of means. Study carried out thanks to Next-generation ThinkinAzul funds (GVA-THINKINAZUL/2021/024).

Results: A significant effect of the developed feed was observed on the essential and non-essential elements on the gilthead sea bream fillets. The highest content of Ca and K was detected in fish feed by PAP (1334 and 15537 mg/kg dw, respectively). On the other hand, it is essential to highlight that no significant differences in t-As content were found between fishes fed by PAP and ALT (647 and 639 $\mu\text{g t-As/kg dw}$, respectively), being the greatest amount of t-As detected in fishes fed by FM (2395 $\mu\text{g t-As/kg dw}$). Taking *Sparus aurata* consumption per capita in Spain in 2022 (0.67 Kg) into account, estimated t-As intake would be 1605 μg per year if feed was FM, being 3.70 and 3.75-fold more if PAP and ALT diet were considered.

Conclusion: The search and use of protein alternatives in the feeding of farmed fish, specifically in gilthead sea bream such as insects, could be a good change to reach a more sustainable and competitive EU aquaculture.

Indigenous and commercial yeasts in Sauvignon blanc wines: growth kinetics and volatile compounds profile

Cássia Cristina Savi², Dr Marcos Roberto Dobler Stroschein², Dr Carolina Pretto Panceri², **Dr Stefany Arcari**^{1,2}

¹Federal Institute Of Santa Catarina - IFSC, ²Federal Institute Of Santa Catarina - IFSC

Aim: Wine quality is influenced by the composition of the grapes and by the microbial communities present during the fermentation process. Aroma is one of the most important attributes for the wine's quality and determine its market value. The aim of this study was to investigate the growth kinetics of *Saccharomyces* yeasts selected from vineyards of the "Vinhos de Altitude" region, compared to commercial *Saccharomyces cerevisiae* yeasts, during the vinification of Sauvignon blanc must and to evaluate the effect of fermentation on the concentration of volatile compounds.

Method: Sauvignon blanc grapes from São Joaquim, Santa Catarina, Brazil (latitude -28°, 14' 9.7", longitude -50°, 4' 16.7", altitude 1,200 meters above sea level) were harvested with 22 °Brix, de-stemmed, pressed and the must was clarified. The control experiment (C) was fermented with strains of *Saccharomyces cerevisiae* (Zymaflore® CX9, Laffort, France). Experiment A was fermented with five indigenous strains of *Saccharomyces*, (01PP, 12M, 13PP, 26PP and 41PP) isolated from leaves and bunches of grapes from vineyards in the "Vinhos de Altitude" region. The fermentation process was monitored by yeast counts in YPD medium (CFU/mL). The determination of volatile compounds was carried out by gas chromatography with a flame ionization detector (GC-FID).

Results: The population of *Saccharomyces* and *Saccharomyces cerevisiae* increased immediately, showing no observable lag phase. The growth curves developed exponential and stationary phases, whose growth kinetics can be described by a three-parameter logistic model. Higher specific growth rate was observed for indigenous yeasts ($\mu_{max} = 4.18$ for A and $\mu_{max} = 2.65$ for C) during the fermentation of Sauvignon blanc wines. The wine fermented with indigenous *Saccharomyces* (A) showed a higher concentration of ethyl hexanoate ($50.53 \mu\text{g.L}^{-1}$) and α -ionone ($2.16 \mu\text{g.L}^{-1}$), while the wine fermented with commercial *Saccharomyces cerevisiae* (C) stood out for higher concentrations of ethyl butanoate ($28.39 \mu\text{g.L}^{-1}$) and phenylethyl acetate ($29.61 \mu\text{g.L}^{-1}$).

Conclusion: Based on experimental evidence, indigenous *Saccharomyces* and commercial *Saccharomyces cerevisiae* are efficient for fermenting white wines. Indigenous *Saccharomyces* produce fruity green apple and violet floral aromas in Sauvignon blanc wines, while commercial *Saccharomyces cerevisiae* yeasts produce rose and honey aromas.

Development of yeast-laden hydrogels for use in 3D-printing systems for the fermentation of alcoholic beverages

Mr Charlie Bailey¹, Dr Thomas Mills¹, Dr Tim Overton¹

¹University of Birmingham

Aim:

Continuous fermentation of alcoholic beverages such as beer has been a desirable outcome for brewers due to reduced costs and improved efficiency. However, beers produced by continuous fermentation are often inferior due to the imbalances in aromatic compounds caused by immobilisation and reductions in mass transfer compared to batch-process beer. The overall aim of this research is to use a 3D-printing system to create yeast-laden bioinks which can be used in continuous fermentation to produce beer with an improved sensory profile over other continuous fermentation systems.

Method:

Different hydrocolloid formulations were printed as bioinks with varying concentrations of freeze-dried yeast. Bioinks were assessed in terms of extrudability, and the ability to create 3D self-supporting structures. The effect on cell viability through printing with different needle gauges was also assessed through live-dead staining. Cast cylinders of 7% Sodium alginate 8% gelatin and 2% carrageenan 8% gelatin, with 1% yeast loading were added to sealed YEPD media flasks to measure fermentation rates. Compression testing was performed to assess the degradation of hydrogels in media over time, as well as the effects of different yeast concentrations.

Results:

Freeze-dried yeast granules were shown to improve the printability of hydrocolloids especially when both were present at higher concentrations. Higher yeast concentrations led to a blending effect whereby the yeast became homogenised during printing, resulting in bioinks with a paste-like consistency. Compression testing showed no clear trend in hydrogel degradation within media, however, increased yeast concentrations resulted in reduced hardness of hydrogels. Alginate-gelatin demonstrated higher rates of fermentation than carrageenan-gelatin and freely-suspended yeast. Both gels showed similar or increased performance during the second fermentation, however, freely-suspended yeast had a lower fermentation rate compared to its initial fermentation. Yeast viability decreased during printing however the different needle gauges did not reduce viability significantly.

Conclusion:

Printing hydrocolloid formulation with freeze-dried yeast appears to be an effective method in adjusting the printability of 3D structures however this comes at the cost of gel strength. Balancing the yeast to hydrocolloid ratio should yield a printing formulation that is both printable and robust enough to withstand use during continuous fermentation.

Autochthonous LAB Strains to Manage Vegetable Fermentation: Study of Leek Fermentation at Low Salt Concentrations

PhD Shuyana Deba¹, Unai Aguirre-Cano¹, Ane Olañeta-Jainaga¹, Iratxe Olazaran-de la Peña¹

¹Leartiker

Aim:

Health consciousness had led to investigate the reduction of sodium in fermented food, which presents biotechnological challenges: improve process control, product quality, and shorten process time. The aim of this study was to characterize the progression of spontaneous leek fermentation based on the salt concentration added, aiming to isolate autochthonous strains of lactic acid bacteria (LAB) for use as starter cultures in controlled low-salt fermentations to reduce the overall fermentation time.

Method:

Organic leeks were purchased from local market; the green and white parts were cut into pieces, rinsed with water, and fermented in brine separately. Two series of spontaneous fermentation were performed in vacuum-packed bags in duplicate with 2 and 5% (w/w) of added salt. Sampling was done in days 0, 1, 2, 3 and 7. A multifaceted methodology was employed: LAB colony counts and isolation, pH and metabolite target transformation. Afterwards, selected LAB were inoculated in 2% brine to validate the application as starter culture for controlled fermentation.

Results:

Regarding spontaneous fermentation, initial pH values were >5.7 which maintained for 24–48 h, then major acidification took place in 2% samples, ending in both green and white parts below 4.2; while 5% white samples remained near 5.5. The microbial community dynamic profile was similar for white and green parts, initial LAB counts were log 3-4, however in lower concentrations bacterial grew significantly faster corresponding with the faster acidification. Day 7 counts were similar in all samples indicating LAB predominance. Glucose and fructose were the main carbohydrates found initially and consumed, ending in lactic acid and acetic acid. Inoculated fermentation showed a faster acidification reaching pH values down 4.3 after 48 h since LAB counts was higher from the beginning, meanwhile carbohydrate consumption rate was higher in the inoculated one.

Conclusion:

In spontaneous fermentation, 2% salt brine was adequate to create a selective medium where LABs were predominant. Additionally, inoculating LAB led to a faster fermentation process. Therefore, the approach of selecting wild-type strains from their natural environments as starter cultures could be an interesting industrial strategy to accelerate the process in a controlled way.

Ecology and technological-functional characterization of wild lactic acid bacteria and yeast isolated from different sourdoughs

Ph.D Iñaki Diez-Ozaeta¹, **Ph.D. Olaia Estrada**¹, Leire Ipiña¹

¹BCC Innovation, Technological Center in Gastronomy, Basque Culinary Center

Aim: Sourdough (SD) fermentation is a traditional biotechnological process used to improve the properties of baked goods. Nowadays, SD fermentation is studied for its potential health effects, such as bioactive compounds and functional microorganisms. Indeed, the aim of the present study is the isolation and characterization of wild lactic acid bacteria (LAB) and yeast to assess their suitability for the production of novel fermented products.

Method: The isolation of LAB and yeast was carried out from five different SD gently provided by artisanal bakery. After isolation and purification of colonies in corresponding culture media, genetic typification through RAPD-PCR and isolate identification was carried out. The characterization of the identified bacteria and yeasts included, among others: i) production capacity of exopolysaccharides (EPS) and vitamin B₂ by LAB; ii) fermentative and acidification ability; iii) enzymatic activities of interest (amylase, protease, phytase); and iv) antioxidant capacity of the strains.

Results: The SD from which isolations were made showed an optimal pH ranging from 3.9-4.3 at the time of plating. After analyzing the counts, it is worth noting a higher proportion of yeast compared to bacteria in white wheat SD (10^7 CFU/mL vs 10^5 CFU/mL), while in SD made with wholemeal flour, populations were found in similar order. RAPD typing revealed a great diversity of profiles, and after sequencing, common species associated with SD, as *Lactobacillus*, *Lactiplantibacillus*, *Leuconostoc*, *Weissella*, or *Saccharomyces* were identified. Then, special emphasis was placed on obtaining vitamin B₂ and EPS-producing LAB. For B₂ production, strains were selected through selective pressure in the presence of roseoflavin. EPS production was assessed after strain growth in the presence of sucrose. Both LAB and yeast strains were evaluated for their leavening and acidification capacity. Additionally, qualitative assessments were made for different enzyme activities, and strain antioxidant capacity in whole wheat extract.

Conclusion: Although further work is necessary to consider other important features, as preservation technology and nutritional-functional properties of fermented products, the study enables the selection of LAB and yeast strains for customize starter cultures for specific requirements, not only for the baking industry but also for the entire agri-food industry.

Artichoke Miso: Fermented pastes as innovative and healthy flavour enhancers

Ms. Idoia Fernandez-pan¹, Mr. Francisco Ibañez¹, Ms Maria Jose Beriain¹, Ms Paloma Virseda¹

¹IS-Food Research Institute. Universidad Publica de Navarra

Aim:

Amino sauces and pastes such as soy sauce and miso are traditional Asian products based on koji (*Aspergillus Oryzae* grown on a starchy substrate) and other raw materials, usually of vegetable origin. Fermented pastes such as miso can be used as flavour enhancers in many preparations due to their high glutamic acid and salt content. Taking into account food trends as well as food reformulation strategies in the industry to reduce the content of additives, and specifically monosodium glutamate, the aim of this work focuses on developing an artichoke miso, rich in umami precursors and reduced salt content, to be used as healthy vegetable flavouring in different food applications such as broths, creams or soups.

Method:

In order to obtain artichoke misos, different formulation, processing and fermentation conditions were evaluated. The rice koji was prepared in a climatic chamber, taking into account as variables the dosage of *A. oryzae* (0.5-1% w/w), time (24-50 h), temperature (25-35 °C) and relative humidity (50-75%). For the preparation of artichoke misos, different rice koji:artichoke ratios were prepared and kept in glass jars at 35 °C for 2 months. The main technological characteristics of the misos were determined, including spreadability tests; their usefulness as a flavouring ingredient in vegetable soups was tested using a 9-point hedonic sensory test to determine their acceptance.

Results:

The resulting conditions for the production of rice koji were as follows: inoculation of 1% (w7w) *A. oryzae* spores over cooked rice and maintenance for 48 h at 35°C and 75% RH. For the artichoke misos, a 1:1 ratio of artichoke to rice koji including 4% salt was preferred. The addition of artichoke lowered the pH of the miso compared to the controls made only with rice. It also increased its firmness and decreased its spreadability. Consumers accepted the use of all the developed misos as vegetable soup flavourings, and preferred the artichoke-containing samples over the control rice misos.

Conclusion:

The use of innovative amino sauces have great potential to be used as ingredients to enhance, promote and facilitate the substitution of flavor enhancers like monosodium glutamate in food products.

Enzymatic Synthesis of Novel Oligosaccharides from Raffinose Oligosaccharides

Philipp Garbers¹, PhD Gordon Jacob Boehlich¹, PhD Jane Wittrup Agger², PhD Birgitte Zeuner², PhD Bjarne Westergaard¹

¹Norwegian University Of Life Science, ²Technical University of Denmark

Aim

Pulses, such as peas, are commonly used as a base for plant protein concentrates and isolates which are a crucial ingredient in meat-alternative foods. However, they also contain a large amount of raffinose oligosaccharides (RFOs) that can reduce the protein percentage in protein concentrates and are classified as fermentable oligo-, di- and monosaccharides and polyols (FODMAPs), which can result in flatulence and gut-related symptoms for consumers. To mitigate this effect, it is beneficial to make less fermentable carbohydrates or enhance their specificity for beneficial gut bacteria. There are some examples of this approach in literature with different enzymes and oligosaccharides hinting at this potential. A similar approach is used in the production of human milk oligosaccharides (HMO), known prebiotics derived from lactose via enzymatic synthesis.

Method

In this study, RFOs isolated from pea and faba bean protein concentrates, pure oligosaccharides, and derivatives (e.g., melibiose) were used as acceptors in transglycosylation reactions with a thermostable β -galactosidase from HMO production with lactose as donor. The reactions and obtained products were characterized by chromatography (HPLC), mass spectrometry (MALDI) and nuclear magnetic resonance spectroscopy (NMR).

Results

Novel oligosaccharides (tri-octamers, MALDI) could be obtained during a variety of reactions with lactose and RFO related oligosaccharides. It could be shown that the addition of melibiose to reactions with lactose and β -galactosidase improved not only the conversion of lactose, but also the observable rate of transglycosylation compared to the hydrolysis reaction. Furthermore, low enzyme loadings ($\leq 0.5 \mu\text{M}$) and short reaction times ($\leq 60 \text{ min}$) benefited transglycosylations. The addition of RFO related oligosaccharides furthermore led to the detection of different transglycosylation products than pure reactions with lactose according to NMR and HPLC.

Conclusion

The desired transglycosylations could be shown and promising new oligos with mixed α - and β -linkages were obtained. These products are hypothesized to reduce fermentability in microbes commonly found in the upper intestinal tract, which could reduce symptoms like flatulence.

The fermentability will be tested in the next steps after a reaction scale-up onto a liter scale membrane reactor to demonstrate industrial feasibility and generate enough material for fermentation studies.

Prebiotic activity of *Monascus purpureus* and *Ganoderma lucidum* mycelia and filtrates towards *Lactobacilli* and *Bifidobacteria*

Chrysanthi Mitsagga¹, Paraskevi Bouki¹, **Dr Ioannis Giavasis¹**

¹University Of Thessaly, Department of Food Science and Nutrition

Aim

Ganoderma lucidum and *Monascus purpureus* are two medicinal fungi that have been used in Asian nutrition and traditional medicine, due to their bioactive compounds, such as pigments, monacolins, γ -aminobutyric acid, polysaccharides, triterpenoids, sterols, proteins and peptides, which are responsible for numerous biological activities, including antioxidant, antitumor, anti-inflammatory, hypolipidemic, hypoglycemic activities. However, their prebiotic potential has not been thoroughly studied. In this study, mycelium and filtrates of *Monascus purpureus* and *Ganoderma lucidum* submerged cultures, as well as “red rice” (produced at lab-scale on solid medium) and *Ganoderma lucidum* commercial powder (grown on solid medium) were comparatively studied for their potential prebiotic activity.

Method

0,1%, 0,5% and 1% from each powder were supplemented to synthetic medium and inoculated with probiotic *Lactobacillus* species (*Lactobacillus rhamnosus*, *L.plantarum* and *Lactobacillus acidophilus*) and *Bifidobacterium* species (*Bifidobacterium breve*, *Bifidobacterium longum* and *Bifidobacterium animalis*) and incubated for up to 72h. The optical density measurements of the probiotic cultures and their colony counts (cfu/ml) with and without addition of fungal supplements were recorded in order to determine the prebiotic activity. The fungal supplements that showed potential prebiotic activity (increase of probiotic populations) were used in in-vitro tests of survival of the *Lactobacillus* and *Bifidobacterium* species in simulated gastric and intestinal fluids. Also, the cell aggregation and eukaryotic cell attachment capacity of the above probiotic bacteria in the presence of the fungal supplements was studied microscopically, based on the attachment of probiotic cells on the cell wall of co-cultured *Saccharomyces cerevisiae* yeast cells.

Results

The addition of *Monascus purpureus* and *Ganoderma lucidum* mycelium and filtrate, as well as “red rice” and *Ganoderma lucidum* commercial powder in synthetic media significantly affected the growth of *Lactobacillus* and *Bifidobacterium* species, either by stimulating the growth of some probiotic species, or by improving the survival rate in gastric and intestinal fluids, at different optimal concentrations of fungal supplements. Attachment to eukaryotic (yeast) cells was also improved in some cases in the presence of fungal mycelium or filtrates. Although the results varied between different probiotic strains, these fungal supplements appear to have significant prebiotic properties, via different mechanisms of action.

Optimization of biomass production and crude protein content of Basidiomycota on carrot side streams

Leonie Juhrich¹, Iris Lammersdorf¹, Lars Tasto², Kai Reineke³, Denise Salzig², Holger Zorn¹, Martin Gand¹

¹Justus Liebig University, ²University of Applied Sciences Mittelhessen, ³GNT Europa GmbH

Aim:

The expanding market for meat alternatives with high protein content includes an appealing option: mycelia, which possess significant nutritional value due to their considerable contents of protein, fibre, and vitamins. Furthermore, industrial side-streams can be used for cultivation, leading to a lower carbon footprint and less waste. After an initial screening, ten fungi/medium combinations were selected for cultivation in media containing two different carrot side streams; response surface methodology (RSM) was employed to optimize the dry biomass (DBM) and crude protein (CP) contents.

Method:

A design of experiment consisting of 34 runs in shaking flasks was conducted for each fungus-medium combination, with pH-values ranging from 3 to 9 and carbohydrate contents varying from 0.4 to 5.4% (black carrot) or 0.6–6.7% (orange carrot). Pre-cultures were grown in 2% malt extract for seven days at 24 °C and 150 rpm in the dark. Subsequently, 100 mL of the side stream medium, supplemented with 10% homogenized inoculum were used for the main cultures. Cultivation was conducted at 24 °C with shaking at 150 rpm in the dark for varying durations, predetermined through a screening for 10 days. For the determination of DBM, mycelia were centrifuged and lyophilized. The CP content was determined using the Kjeldahl method with a nitrogen to protein conversion factor of 4.5.

Results:

Using RSM, the pH value and carbohydrate content were optimized to maximize the yields of DBM and CP for all fungus-medium combinations. These optimized conditions were then validated through a three-fold validation process. While the DBM content showed significant improvement, the effect on CP was less evident.

Conclusion:

Compared to screening conditions (orange carrot: pH 4.20, carbohydrate content 2.2%; black carrot: pH 3.55 and 1.8% carbohydrate content), optimization of all parameters could be achieved. For example, DBM increased by a factor of 2.6, CP content by a factor of 2.1. Thus, mycelia cultivated on carrot side streams emerge as a promising protein source for vegan nutrition, with a yield that can be further enhanced through optimization of various cultivation parameters in bioreactors.

Inoculation of endophytic fungi into "Throuba" olives for natural de-bittering under controlled conditions

Dr Sofia Chanioti¹, Panagiota Stergiou¹, Zacharoula-Maria Xanthou¹, Ioanna Chalvantzi¹, Dr Aspasia Nisiotou¹, **Sofia Chanioti**¹

¹Institute of Technology of Agricultural Products ELGO-DEMETER

Aim:

The aim of this study was the endophytic fungi isolation from "Throuba" olives and the inoculation of the fungal isolates through different techniques into unripened "Throuba" olives for natural de-bittering under controlled conditions.

Method:

Ripened "Throuba" olives growing on Heraklion, Crete were collected and subjected to endophytic fungi isolation by using malt extract agar (MAE). DNA was extracted from fungal cultures, and PCR was performed for the amplification of the ITS-5.8S rDNA region using the primers ITS1 and ITS4. PCR products were purified and directly sequenced. To obtain suspensions for olive inoculation, different fungal isolates were cultured on MEA and incubated at 30°C in the dark. After 10 days, conidia were collected and transferred to 10ml of 0.01% (v/v) Tween 80 adjusting a concentration to 10⁶ CFU/ml. Unripened "Throuba" olives were inoculated with the fungal suspensions either by immersion for 24 and 96h (Fungus-Im-24 and 96) or by spraying (10 sprays in 15s-Fungus-Spray). The inoculated olives were placed in sterile dishes and incubated at 25°C for 20 days. Inoculated "Throuba" olives were evaluated for their natural de-bitterness in terms of oleuropein content and their quality characteristics (moisture, color, texture, total phenolic content-TPC, antioxidant activity, total anthocyanins, β -glucosidase and polyphenol oxidase activity) at various time intervals (0, 4, 11 and 18 days of storage).

Results:

Fungus-Im-24 and 96 resulted in surface change color from purple to black during storage. At 20 days of storage, Fungus-Im-96 had the lowest hardness values, followed by Fungus-Spray olives and Fungus-Im-24. The TPC of inoculated "Throuba" olives decreased from 32.35 to up to 3.94 mgCA/g on the 20th day of storage. The lowest values of the TPC were observed in Fungus-Im-96 olives, followed by Fungus-Im-24 and Fungus-Spray ones. The concentration of oleuropein in Fungus-Im-96, Fungus-Im-24 and Fungus-Spray olives was reduced by 96.92%, 92.89% and 82.48%, respectively, at the end of the storage indicating that immersion for 96h accelerated olives' de-bittering process.

Conclusion:

Endophytic fungal isolates could be used for the inoculation of olives allowing for their natural de-bittering under controlled conditions, thus establishing the comparative advantages of the application of standardized debittering over physical.

Potential of exopolysaccharide-producing starter cultures in vegan coconut yoghurt analog manufacture

Ms. Sophie Libberecht¹, Mia Ristevska¹, Myriam Loeffler¹

¹KU Leuven - Dept. of Microbial and Molecular Systems; Research Group MTSP

Aim:

To meet consumer demands in terms of health promoting effects and "cleaner" labels, while at the same time offering products with a pleasant texture and taste, the food industry is constantly seeking solutions to further improve dairy analog quality. An interesting approach could be the use of exopolysaccharide (EPS)-producing lactic acid bacteria (LAB), as EPS are known for their gelling, water-binding, thickening, and texturizing effects. In addition, the EPS formed *in-situ* do not have to be declared. The aim of this study is to investigate the potential of an EPS forming *Lactobacillus sakei* strain in the manufacture of a vegan protein-enriched coconut yoghurt analog without the use of other hydrocolloids/thickeners.

Method:

Vegan yoghurt analogs were prepared by heating coconut milk containing 5% w/w added sucrose, no or added pea protein isolate (PPI; 0-5% w/w), and 0-1.5% w/w tapioca starch, to 90°C for 3 min. After cooling, the mix was either inoculated with the EPS-producing strain *L. sakei* 1.411 or with the non-EPS-producing control *L. sakei* 1.2037 (10⁶ CFU/g) and the analogs fermented at 30°C until a pH of 4.5, followed by storage at 4°C. Microbial growth and pH were monitored throughout the experiment. *In-situ* formed EPS were quantified using HPLC and firmness (backwards extrusion) as well as susceptibility to syneresis were assessed after 1 and 5 days of storage. Moreover, a descriptive sensory evaluation was performed.

Results:

Independent of the product composition and strain used, the pH of the products decreased from pH 6.36±0.16 to pH 4.47±0.13 over the course of fermentation with anaerobic cell counts reaching approx. 1.00 x 10⁹ CFU/g after 18 h of incubation. Significant differences (p<0.05) could be detected between samples containing 5% PPI but no starch. Here, only samples containing *L. sakei* 1.2037 showed syneresis (12.57±0.52%). In contrast to the strains used, the presence of starch had a pronounced effect on the firmness of yoghurt analogs containing 5% PPI. However, the samples with *L. sakei* 1.411 achieved better results in the internal sensory analysis.

Conclusion:

The production of yogurt analogs with EPS-forming LAB seems to be a promising approach that will be further examined.

Optimization of the sensory overall impression of a fermented egg white product

Insa Mannott^{1,2}, Erik Schledermann¹, Prof. Dr. Ramona Bosse¹, Prof. Dr. Monika Gibis²

¹Department of Food Technology, Institute EcoMaterials, University of Applied Sciences Bremerhaven, ²Department of Food Physics and Meat Science, Institute of Food Science and Biotechnology, University of Hohenheim

Aim:

In recent years, the trend toward protein-rich diets has resulted in the development of new protein-based products. The focus has shifted to protein-enriched desserts, as taste and enjoyment remain the most important decision criteria. In addition to plant-based proteins, egg white products offer a sustainable and resource-conserving source of protein. This source can be used to create innovative desserts. The egg white is by nature lactose-free, rich in protein, and can be processed in a sensory and microbiologically valuable with a fermentation process. The challenge of this study was to advance the product development of this innovative dessert and to develop marketable flavors for the fermented egg white base that would lead to consumer acceptance.

Method:

In this study, a basic recipe for an egg white dessert was developed based on the existing egg white base. The parameters sweetness and aroma were optimized. The sensory acceptance was evaluated by an internal, trained panel (n= 7 to 15) based on the Just-About-Right method (JAR scale, five levels). In addition, a consumer test (n = 77) was conducted with randomly selected passers-by in Bremerhaven to analyze the acceptance of the product. Acceptance was measured on a seven-point scale with a total of seven questions per sample.

Results:

An initial test series with four concentrations from no sweetness to high sweetness was created and analyzed by the internal panel to optimize the sweetness of the innovative egg white base. The results showed that the sweetness could be significantly reduced compared to market-standard desserts, thus contributing to the sugar reduction strategy. In an external consumer test, the dessert variant was optimized for sweetness, taste, and color and evaluated for overall acceptability. The dessert was found to be very good (19%) to good (38%) overall consumer acceptance.

Conclusion:

In conclusion, this study shows that appealing and marketable flavors could be developed for the innovative egg white dessert. This means that by-products from the processing of egg products can also be used as sustainable and resource-saving protein sources for sensorially appealing dessert products and open up new sources of added value.

Use of phytase-active *Pediococcus pentosaceus* in sourdough fermentation to enhance the bread quality

Dr. Matin Mohammadi Kouchesfahani¹, Zohreh Hamidi-Esfahani¹, Mohammad-Hossein Azizi¹

¹ Department of Food Science and Technology, Faculty of Agriculture, Tarbiat Modares University, Tehran, Iran

Aim:

Employing wheat bran in bread-making is acknowledged as a way to provide carbohydrates, proteins, dietary fiber, and bioactive compounds and decrease the risk of cardiovascular disease, gastrointestinal cancer, and type 2 diabetes. Although phytic acid in wheat bran as an anti-nutritional factor is a common concern. Phytic acid can form complexes with Fe⁺², Zn⁺², Ca⁺², Mg⁺², Cu⁺², and Mn⁺². Furthermore, the addition of bran has detrimental effects on the technological and rheological characteristics of bread.

Method:

In this study, fermentation with phytase active *Pediococcus pentosaceus* mk.zh 95 was employed to reduce the negative effect of bran addition. Sourdough was added to bread samples at a level of 15% (w/w). Moreover, in all samples, wheat flour was substituted with wheat bran at the levels of 10 and 15% (w/w).

Results:

Fermentation with *Pediococcus pentosaceus* mk.zh 95 resulted in a decrease in phytic acid content. Additionally, bread samples containing *Pediococcus pentosaceus* mk.zh 95 had higher acidification rate, moisture content, and lower hardness compared to the control samples.

Conclusion:

Our results showed that *Pediococcus pentosaceus* mk.zh 95 could be employed as a starter culture in bread-making to enhance the nutritional value of bread.

Exploiting RNA thermometers (RNAT) as molecular control element for precision fermentation

Christina Peterzell¹, Philipp Noll¹, Marius Henkel¹

¹Cellular Agriculture, TUM School of Life Sciences, Technical University of Munich

Aim:

In the recent past, an increased interest in alternative protein sources has led to a rethinking of protein from fermentation processes. For food applications, either as bulk protein or functional additive, more efficient processes are required to be cost-effective. A neglected starting point for increasing the efficiency of bioprocesses is the decoupling of biomass and product formation. In most cases, biomass is a by-product that is difficult to recycle efficiently. For this task, RNA thermometers (RNAT) can be exploited as a gene expression 'dimmer switch'. RNATs adopt a hairpin-like structure that masks the ribosome binding site (RBS) affected by temperature changes leading to RBS exposure and translation initiation.

Method:

Plasmids containing fourU-RNAT and EmGFP as a reporter gene are constructed in *E. coli*. Biomass growth (optical density), substrate consumption (enzymatic assays) and gene expression (fluorescence assay) are recorded over temperature range of 25-40 °C. The optimal balance between substrate conversion for protein expression and growth is identified by calculating key performance indicators e.g. yields (substrate conversion (g/g)) and rates (g/g h). Subsequently, temperatures optimal for growth are selected and temperature shifts between these optima are performed. So-called off-on-off switches are carried out in subsequent experiments to test the natural reversibility of the induction system described in literature.

Results:

Obtained data and characteristic process parameters are the basis for the establishment of a descriptive process model. As a result, the optimum temperature range can be further minimized, allowing a highly accurate prediction and determination of the established RNAT-based expression system. To date, off-on-off switches are in the stages of experimental trials and hold promising potential for future applications.

Conclusion:

Hence, the expression platform in combination with temperature changes can be used to direct the carbon substrate into product rather than (waste) biomass optimizing the conversion yield and thereby process efficiency. Therefore, molecular bioprocess control exploiting RNAT in combination with temperature changes represents a valuable tool for enhanced efficiency of future food precision fermentation processes.

Exploring the use of hydrocolloid gums in goat cream cheese production

Dr Isabel Ratão^{1,2}, Dr Patrícia Nunes^{1,2}, Vanessa Silva¹, **Dr Célia Quintas**^{1,2}

¹Universidade Do Algarve, ²Mediterranean Institute for Agriculture, Environment and Development & CHANGE–Global Change and Sustainability Institute, Faculty of Sciences and Technology, University of Algarve

Aim: The Mediterranean region's most popular dairy products are yogurts and cheeses, particularly those made from goat's and sheep's milk. The most traditional dairy products in southern Portugal are goat's and sheep's cheeses. This study aimed to produce cream cheese with adequate texture and spreadability by selecting the best hydrocolloid gum, in order to diversify the products made from goat's milk from the Algarvian breed.

Method: The cheese was processed with goat's milk, salt (12 g/L), calcium chloride (0.5 g/L), and a coagulant of vegetable origin (30 drops/L). Three combinations of gums (0.5% (w/w) were tested: locust bean gum (A1); xanthan (A2) and (locust bean gum: 1 carrageenan) (A3). The quality of the cream cheese was evaluated through the study of microbial [aerobic microorganisms (AM) (30°C), fungi, coagulase-positive staphylococci, and *Escherichia coli*] and physicochemical (yield (%), pH, a_w, and fat (%)) parameters.

Results: Coagulase-positive staphylococci, *E. coli*, and molds were not detected in the cream cheese. However, yeast numbers increased from 1 log cfu/g to about 2 log cfu/g on the fourth day of storage and AM (30 °C) enumeration was higher in A1 and A3 (approximately log 3 cfu/g), with a slight increase on day 4. Regarding A2, log 1 cfu/g was found with almost the same value on day 4. The pH showed no significant variation among the cheeses studied (6.49±0.032) but a very slight decline (6.42±0.05) over the storage period was observed. No significant differences existed between the cheeses or over the storage period (4 days) in the a_w values (0.963±0.004). The fat content in the cheeses was 12.0±0.0%, 12.1±0.3%, and 11.8 ±0.4%, respectively in A1, A2 and A3.

Conclusion: The cream cheeses under investigation exhibited comparable properties, with an estimated shelf life of approximately four days. Furthermore, according to the regulation of the European Commission (EC, 2005, EC, 2007) the cheeses studied satisfied the microbiological criteria for food hygiene. However, the best spreadability was obtained when locust bean gum was added to the cheese.

Balancing aroma generation and food safety in vegan raw-fermented sausage analogues via pre-fermentation

Soramony Sam¹

¹University Of Hohenheim

Aim:

Due to naturally high carbohydrate contents of plant materials, vegan raw-fermented sausage analogues obtained via microbial fermentation with lactic acid bacteria (LABs) can result in very low pH values, which negatively affects sensory properties. However, microbial fermentation is crucial for generating desirable aromatic compounds, and for providing food safety. Therefore, a partial two-stage fermentation was postulated in which a fraction of the raw materials should be pre-fermented in a controlled environment before the actual product was manufactured. Hence, low-moisture extruded protein texturates (LMPT) were fermented with varying microorganisms at a range of fixed a_w -values, adjusted via NaCl. The fermentation was assessed regarding development of pH and aroma, as well as microbial safety.

Method:

Wheat LMPT was soaked in brine (6 %, 8 %, 10 % NaCl) and inoculated with different combinations of microorganisms: a) *L. sakei*, *Z. bailii*, *S. carnosus* (= LYS); b) *Z. bailii*, *S. carnosus* (= YS); c) *Z. bailii* (= Y); d) *S. carnosus* (= S). Fermentation was performed at 15 °C for 8 days. pH and a_w were recorded at regular time intervals, bacterial cell counts (cfu/g) and volatile compounds (GC-MS) were recorded at $t_0 = 0$ days and $t_{end} = 8$ days.

Results:

Fixed a_w -values (0.955, 0.933, and 0.906 for 6 %, 8 %, and 10 % NaCl, respectively) remained constant over time. pH values slightly decreased in LYS, YS and Y at 6 % NaCl (= LYS-6, YS-6, Y-6). Bacterial growth was found in all samples with 6 % NaCl, during which cell counts increased by up to two orders of magnitude. In Y-8 and Y-10, yeast cell counts decreased by one order of magnitude. In LYS, YS and S at 8 – 10 % NaCl, the populations of LABs, staphylococci and yeasts remained relatively constant. (*further results pending*)

Conclusion:

At >6 % NaCl, yeast populations displayed higher survivability in presence of *S. carnosus* and/or *L. sakei*, indicating synergistic interactions. Therefore, we recommend combining *Z. bailii* and *S. carnosus* in pre-fermentation of wheat LMPT at 6 % NaCl or higher. For food safety reasons, we also suggest to additionally include *L. sakei*.

Antioxidant properties of cereal flours fermented with selected microorganisms

Phd Student Elena Tomassi¹, Agnese Sgalippa¹, PhD Laura Pucci¹

¹Institute of Agricultural Biology and Biotechnology-CNR

Aim:

Reactive oxygen species are molecules containing one or more unpaired electrons, which can damage nucleic acids, proteins, and lipids, leading to several diseases including early aging, cancer, inflammatory disease, and diabetes. Antioxidants play a crucial role in preventing cellular damage by reducing oxidative stress with beneficial effect on human health. Microorganisms are of great biotechnological interest for their capacity to produce bioactive compounds with antioxidant activity during fermentation processes. This study aimed to assess the impact of fermentation on the antioxidant properties of cereal flours, optimizing time, temperature, and microbial strains.

Methods:

Fermentation was carried out by adding a starter culture containing lactic acid bacteria and yeast strains into a mixture of flour and water. Polyphenols content was evaluated with Folin-Ciocalteu method, while flavonoids content was determined by aluminium-chloride method. DPPH, ORAC and FRAP tests were performed to detect the antioxidant activity and free radical scavenging of fermented flour extracts. Cellular antioxidant activity on red blood cells (CAA-RBC) was conducted to evaluate *ex vivo* antioxidant properties of fermented flour extracts and to provide information on the ability of the compounds to cross the plasma membrane.

Results:

Following the analyses of several cereal flours (whole-wheat, spelt, tef, oat, rice), the optimal fermentation time selected was 4 days, the best temperature was 37°C and the most antioxidant fermented flours was the whole-wheat. Subsequently, the whole-wheat flour was fermented with eight different "MIX" of selected microorganisms to optimize the fermentation process, based on polyphenols and flavonoids content, and DPPH scavenging activity. Particularly, the "MIX 8" fermentation increased polyphenols and flavonoids content compared to the unfermented flour (3.40±0.04 vs 1.22±0.02 mgAGE/g and 2.55±0.17 vs 1.23±0.24 mgCE/g respectively) and enhanced the antioxidant activity *in vitro* both in DPPH (0.92±0.06 vs 9.47±0.59mg/ml), ORAC (1110.21±27.81 vs 3521.24±42.85 µmolTE/100g) and FRAP (84.50±0.71 vs 811.17±12.96 µM Fe⁺⁺) assays. Moreover, the incubation of RBCs with fermented and unfermented flour extracts, under prooxidative condition, showed an improvement in the antioxidant capacity of "MIX 8" (56.43±5.80 vs 28.99±3.73 CAA unit, respectively).

Conclusions:

The fermentation process, with a selection of microbes, temperature and timing, significantly implemented the antioxidant properties of wheat flour.

Impact of Reaction Flavor on Biogenic Amines and Savory Aroma in Anchovy Sauce

Su-Min Kim¹, Kyeong Hwan Hwang¹, Jeong Min Heo², Youg Hee Jung², Jeong Jin Seo², Changheon Lee¹, Yong-Jun Cha², Daeung Yu^{1,2}

¹Interdisciplinary Program in Senior Human Ecology, Major in Food and Nutrition, Changwon National University, ²Department of Food and Nutrition, Changwon National University

Aim: The aim of the study was to develop anchovy seasoning sauce with savory, bulgogi, and umami flavors while utilizing reaction flavor with specific amino acids (threonine, glutamic acid, and glycine). The goal was to address the declining preference for anchovy sauce in the industry due to its distinct odor and off-flavors, as well as the issue of exceeding biogenic amine content standards in advanced countries. The research focused on leveraging precursor materials to improve the fermentation process of anchovy sauce, aiming to concurrently enhance preferred aromas while reducing biogenic amine content.

Method: Utilizing amino acids (threonine, glutamic acid, and glycine) as precursor materials, reaction flavor technology was applied during anchovy sauce fermentation to reduce biogenic amine occurrence and enhance preferred aromas, followed by sensory evaluations on a 9-point scale to assess consumer preferences, alongside adjustment of salt concentration below 20% to mitigate excessive salt content typical in commercial products.

Results: The study successfully enhanced consumer preference by over 53% compared to commercial products, integrated prototypes into industrial processes, adjusted salt concentration below 20% to mitigate excessive salt content, and effectively reduced biogenic amine occurrence using reaction flavor technology.

Conclusion: The successful development of anchovy seasoning sauce using reaction flavor technology addressed industry challenges, resulting in products with increased consumer preference and added value, potentially offering health benefits through reduced biogenic amine content and optimized salt concentration for enhanced antihypertensive and physiological functionalities.

Production and Characterization of rLysEc301, The Lytic Enzyme of *E. coli* Bacteriophage

Student Aysenur Yücefaydalı¹, Prof. Dr. Yeşim Soyer¹

¹Middle East Technical University

Aim:

The increasing incidence of antibiotic resistance in *E. coli*, a common pathogen causing foodborne illnesses, has led to a rise in hospitalization cases. As a result of the resistance of bacteria to antibiotics and bacteriophages, it is obvious that a new way must be found in order to minimize the diseases caused by contaminated food in the food industry. Lysins, the lytic enzyme of bacteriophages, may be a promising solution. Therefore, the aim of this study is to produce lysin enzyme using recombinant DNA technology and purify lysin and to investigate the inhibitory effect of lysin on bacteria.

Method:

In this study, the genome of bacteriophage MET P1-301, specific to *E. coli* O104:H4, was sequenced using Illumina and Oxford Nanopore platforms. A putative lysin, LysEc301, was identified and cloned into *E. coli* BL21 using a pET-28a(+) vector. LysEc301 was expressed and purified using the Thermo Scientific HisPur™ Ni-NTA Purification Kit. Experiments were conducted to assess the lytic activity of recombinant LysEc301 (rLysEc301) against *E. coli* strains O104:H4 and O157:H7 in various conditions, including in milk.

Results:

Recombinant LysEc301 demonstrated high lytic activity, reducing *E. coli* concentration by approximately 3 log(CFU/mL) in lytic activity test. It was effective both individually and synergistically with EDTA. The yield of recombinant LysEc301 was approximately 100 ng/μL, and the protein exhibited a molecular weight of 17 kDa. Its antibacterial effects were confirmed in both buffer solutions and milk environments, showcasing its potential as a biocontrol agent in the food industry.

Conclusion:

The study confirms the effectiveness of rLysEc301, a lytic enzyme from bacteriophage MET P1-301, in significantly reducing *E. coli* levels, offering a promising alternative to antibiotics. The results suggest that lysins can be integrated into food safety protocols to combat antibiotic-resistant bacteria, potentially transforming biocontrol strategies in the food industry.

Lactic acid bacteria as technological and sensory biomodulators for novel plant-based fermented beverages

Ph.D Iñaki Diez-Ozaeta¹, Olaia Estrada¹, Telmo Puente¹, Laura Vázquez-Araújo^{1,2}, John Regeffalk^{1,2}
¹BCC Innovation, Technology Center in Gastronomy, Basque Culinary Center, ²Basque Culinary Center, Faculty of Gastronomy Sciences, Mondragon Unibertsitatea

Aim: Rising concerns about the environmental impact of dairy farming, ethical considerations, and medical needs, such as lactose intolerance and cows' milk protein allergy, are driving consumers towards plant-based dairy alternatives. The present work aims to gain knowledge into the transformative potential of microorganisms in refining the flavor of plant-based beverages.

Method: The performance of commercial yogurt cultures (CH) and selected LAB strains (LLV) were compared in dairy milk and rice beverage. After the fermentation of both matrices characterization of the developed prototypes was carried out in terms of cell viability, pH, total acidity, total soluble solids, color, aromatic compounds, and sensory perception.

Results: The matrix itself significantly influenced the parameters assessed throughout the fermentation. In fact, higher cell viability ($>10^8$ CFU/mL) and pH reduction ($\text{pH}<4.3$) was observed in rice-based beverages. By contrast, dairy fermented beverages reported approximately 10-fold higher total acidity than rice-based beverages. After the fermentation, both matrices showed a significant change in the ΔE^* (degree of color difference), although specifically the rice drinks, and the LLV-fermented combination, showed a greater visually perceptible shift which may be due to the oxidative degradation of proanthocyanidins of the grain. Volatile composition suggested that the main differences were also matrix-dependent. However, fermentation had a significant role in altering the composition of both dairy milk and rice beverage samples. In rice-based beverage a significant reduction of aldehydes, ketones, and alcohols which are linked to beany/legume off-flavor was reported, by contrast, a significant increase in total acids was confirmed. Sensory analysis also highlighted the influence of the matrix, with rice beverage and its derivatives exhibiting more cereal, legume, nutty-like, and watery characteristics than the dairy products. However, fermentation reduced the typical perception of sweetness and cereal notes of plant-based beverages, while increasing the acidity of fermented drinks.

Conclusion: The results of the analyses showed the clear effect of the raw material on the ongoing fermentative process. Further characterization of the selected bacterial strains holds the key to unlocking fermentation's full potential in mimicking the nutritional, sensory, and textural qualities of dairy products.

Screening and Characterization of Biosurfactant-Producing *Bacillus* Species Isolated from oil contaminated Soils in Tunisia

Prof.dr Zied Zarai, Houda Gharlallah¹, Mecit Halil Öztop

¹University of Sfax, High Institute of biotechnology of Sfax, Food Technology departement, Sfax, Tunisia, ²Middle East Technical University, Faculty of Engineering, Food Engineering, Ankara, Turkiye

The study emphasizes on the production of biosurfactant from different bacillus species isolated from an olive oil-contaminated site of Sfax. Maximum reduction of surface tension was recorded when cultivating isolates in the OM medium composed of (g/L): peptone 20, sucrose 25, yeast exact 4.5, KH₂PO₄, MnSO₄ 0.006, and MgSO₄ 0.6. The cell free supernatant was used to screen the ability of the isolate to produce biosurfactant. Based on hemolytic screening method, four strains, S40, S132, S15 and H6 were selected as potent producers of biosurfactant and molecular sequencing confirm that isolates were related to *Bacillus amyloliquefaciens*, *Bacillus velezensis*, *Bacillus subtilis* and *Bacillus* sp., respectively. Crude biosurfactants was characterized as lipopeptide by FTIR and TLC analysis. Furthermore, a detailed study to characterize the chemical composition of the biosurfactants produced was carried out. Ultra-high-performance liquid chromatography coupled with high-resolution mass spectrometry (UHPLC-MS/MS) revealed the dominance of a series of Surfactin homologues, Utirin and fengycin class were identified.

Keywords: Biosurfactants; emulsification index; *Bacillus*; surface tension; Drop collapse

L. monocytogenes' responses to plasma-processed air decontamination

Domiziana Battaglia¹, Shehreen Ferdous¹, Dr. Masja N. Nierop Groot², Prof. Tjakko Abee¹, Prof. Heidy M. W. Den Besten¹

¹Food Microbiology, Wageningen University & Research, ²Wageningen Food & Biobased Research, Wageningen University & Research

Aim:

Non-thermal plasma is a promising processing technology for surface decontamination as an alternative to conventional methods such as heat treatments and chemical additives. This study explores the impact of plasma-processed air (PPA) exposure on *Listeria monocytogenes*, the effects of strain variability, and the possible resistance mechanisms including secondary oxidative stress response.

Method:

A microwave-driven plasma source was used to produce PPA with atmospheric air as carrier gas. Ten wild-type (WT) strains from different origins and with different resistance profiles were exposed to PPA and their susceptibility was compared. In addition, the *L. monocytogenes* EGDe sigB knock-out mutant strain was screened to assess the role of this general stress response regulator in PPA resistance and also towards peracetic acid-induced oxidative stress. Then, to explore possible cross-protection towards PPA induced by mild oxidative stress, EGDe WT and Δ sigB mutant were exposed to sub-lethal concentrations of peracetic acid followed by PPA treatment. Lastly, transcriptomic analysis was performed to evaluate the genomic response after PPA exposure.

Results:

No strain variability was detected among the ten WT strains, and also the EGDe Δ sigB mutant showed similar inactivation upon PPA treatment. In contrast, the EGDe Δ sigB mutant showed higher sensitivity to peracetic acid exposure compared to EGDe WT, indicating SigB activation and its role against oxidative stress. The mild pre-treatment with the oxidative agent peracetic acid showed a synergistic effect with PPA in the bacterial inactivation of *L. monocytogenes*. Transcriptomic analysis shed light upon up- and down-regulated molecular pathways of PPA-treated cells, indicating which adaptation processes took place.

Conclusion:

The low strain variability in PPA resistance is a promising observation for industrial applications. Although PPA and peracetic treatment showed synergistic effects, SigB seems to play a limiting role in PPA resistance, while it has a role in peracetic acid resistance, pointing to complementary resistance mechanisms in PPA-treated cells.

Antimicrobial activity of PAW against *E. coli* planktonic cells & biofilms formed on polystyrene surfaces

Ms Raquel Camilleri¹, Prof. Ruben Gatt¹, Prof. Vasilis Valdramidis^{1,2}, Dr. Foteini Pavli¹

¹University of Malta, ²National and Kapodistrian University of Athens

Aim:

The aim of the study was to develop bacterial biofilms of *Escherichia coli* NCTC 12900 under selected environmental conditions on a polystyrene surface and to evaluate plasma-activated water (PAW) for its disinfection potential. Furthermore, the disinfection ability of PAW on *E. coli* NCTC 12900 planktonic cells, was also examined. The project's significance is to strengthen the knowledge on the disinfection properties of PAW, and assess its potential as an alternative disinfection method for the food industry.

Method:

PAW was produced at atmospheric pressure using a copper electrochemical cell, and its final pH value was recorded on every sample. *E. coli* NCTC 12900 was used throughout the study, whilst sterile distilled water was used as a control for disinfection. Resistance to PAW was assessed on planktonic cells after exposure for 0, 10, 20, 30, 40, 50, and 60 min. For the biofilm formation, a pre-attachment step was performed for 3h, followed by addition of Tryptic Soy Broth for 48h at 37° C. The biofilms developed on polystyrene were exposed to PAW for 0, 5, 10, 15 and 20 min. Biofilm detachment was performed by cell scraping.

Results:

The results obtained showed approximately a 5 log CFU/mL reduction of planktonic cells after exposure to PAW for a total of 60 min. For the developed biofilm, PAW application for 20 min resulted in a 2.5 log CFU/cm² reduction.

Conclusion:

This study indicates that PAW has the ability to be utilised as a disinfection agent on food contact surfaces, however prolonged contact time is required to achieve significant disinfection rates. Furthermore, additional research is needed to establish whether PAW can be utilised by the food industry as a disinfectant, given that some of its components might exert toxic effects.

Effect of light and drying method on bioactivity of *Monascus purpureus* and *Ganoderma lucidum* cultures

Dr Ioannis Giavasis¹, **PhD Student Chrysanthi Mitsagga**

¹University Of Thessaly, Department of Food Science and Nutrition

Aim:

Ganoderma lucidum and *Monascus purpureus* are two medicinal fungi which produce numerous bioactive compounds. *G. lucidum* produces β -glucans like ganoderan and ganoderic acid, which have anticancer, antioxidant, anti-inflammatory, immunostimulating, hypocholesterolemic, hypoglycemic activities, while *M. purpureus* is known for the synthesis of monacolin, a cholesterol-lowering compound, antioxidant pigments and other bioactive substances. Cultivation and drying conditions were studied in order to optimize each bioprocess.

Method:

The two fungi were cultured in petri dishes, shake flasks, solid medium (rice) and 2L Stirred Tank Bioreactors in previously optimized bioprocess parameters under different types of light (darkness, sunlight, or white, red, blue, green artificial led light). The filtered mycelia and the fermentation broth with all extracellular bioactive metabolites were isolated, dried with either freeze-drying or hot-air (45°C) drying and then extracted as either water or ethanol extracts. All samples were studied for their antioxidant and antimicrobial activity, which were also related to polysaccharide and pigment production. These bioactive properties were compared with (a) commercially available *Ganoderma lucidum* powder from fruiting bodies of the mushroom and (b) red rice with *Monascus purpureus* produced in a solid state fermentation in the lab.

Results:

The results showed that the bioactive (antioxidant and antimicrobial) properties are present in both fungi, either when grown in solid medium or when isolated from submerged cultures. However, different content of phenols and flavonoids exist in different fraction of liquid or solid fermentation and the production of pigments or the age of the mycelium may affect the bioactive properties. The water extracts of the dried mycelium of both fungi were not significantly effective against the tested microorganisms while the ethanol extracts were effective against *E. coli*, *L. monocytogenes*, *S. aureus* and *P. expansum*. Freeze-drying led to improved antioxidant activity compared to hot-air drying. The type of light influenced the growth and bioactive properties of the tested fungi. Red or blue light was the optimal source of light for mycelium growth and antioxidant effect of *G. lucidum*. *M. purpureus* grew well in darkness and in red light, while blue light decreased its growth rate, pigment and phenol content, antioxidant capacity and monacolin content.

Shelf life of peach juice cold pasteurized by pulsed electric fields: comparison with thermal treatment

Sofia Chanioti¹, Dr. Varvara Andreou¹, Dr. Marianna Giannoglou¹, Mrs Zacharoula-Maria Xanthou¹, Dr. Athanasios Limnaios², Mr Aleksandros Katsimichas², Dr. Petros Taoukis²

¹Institute Of Technology Of Agricultural Products Elgo-demeter, ²National Technical University of Athens

Aim:

Pulsed electric field (PEF) technology has emerged as an alternative to traditional pasteurization methods for microbial inactivation in heat-sensitive liquid foods, thereby preserving their physicochemical characteristics, such as color and functionality. PEF induces membrane electroporation, leading to reduced microbial growth rates during storage and extending the shelf-life of liquid products while retaining their organoleptic properties.

This study aimed to assess the potential advantages of applying PEF to preserve peach juice and compare its efficiency to conventional thermal pasteurization.

Method:

Peach juice was thermally (80 - 90 °C for up to 5 min) and PEF (20 kV/cm electric field strength, frequency of 400 Hz, pulse width 15 µs, 250 mL/min flow rate) treated (conditions were selected by previous work on kinetic study of PEF conditions on quality indices of the juice). Following pasteurization, the samples were aseptically packaged in glass containers and stored under refrigeration at temperatures of 5, 10, and 15 °C. Throughout storage, microbial analysis and various quality parameters, including color, pH, viscosity, and organoleptic characteristics, were monitored for both samples at all temperatures studied. Additionally, the concentration of several antioxidant compounds, such as total phenolic compounds and vitamin C was assessed. Shelf-life determination was also conducted and compared for both studied samples.

Results:

Results revealed that PEF processing did not cause any significant change in pH, color (L*, a*, and b*), and viscosity of pasteurized juices. PEF treated juices were comparable to thermal pasteurized juices regarding the efficacy in the inactivation of aerobic bacteria and lactic acid bacteria. Neither PEF nor thermal treatments significantly altered the total phenolic content or antioxidant activity of the juices. Sensory evaluation revealed that PEF-treated juices exhibited a more fresh-like organoleptic profile compared to those pasteurized thermally.

Conclusions:

PEF processing stands out as a practical approach for the pasteurization of fruit juices with fresh like characteristics, while simultaneously ensuring their microbial stability and enhancing their concentration of health-promoting compounds.

Innovative strategies to prevent and eliminate *Listeria monocytogenes* biofilms: Plasma-Polymerized Coatings and Plasma-Activated Water

Ms Ángel Francés¹, Dr Márcia Oliveira¹, Miss Rebeca Cordero-García¹, Dr. Rodolfo Múgica-Vidal², Dr Fernando Alba-Elías², Dr Montserrat González-Raurich¹, Dr Avelino Álvarez-Ordóñez¹, **Dr M.**

Mercedes López Fernández¹

¹University of León, ²University of La Rioja

Aim: Biofilms present in food processing facilities fosters cross-contamination and food spoilage, posing health risks to consumers. Traditional disinfection methods struggle to eradicate biofilms, necessitating the development of new strategies to prevent and remove bacterial attachment to surfaces. Plasma technology has emerged as a promising alternative for decontaminating food processing environments and enhancing food microbiological quality. This includes the potential of anti-biofilm coatings applied with an atmospheric-pressure plasma system and the use of plasma-activated water (PAW), which offer promising approaches for biofilm elimination. This study aims to evaluate PAW's effectiveness in eradicating *Listeria monocytogenes* biofilms on stainless steel (SS) surfaces and to investigate the application of various plasma-polymerized coatings on SS to hinder bacterial adhesion by altering the surface's physico-chemical properties.

Method: Coated and uncoated SS plates were used to grow a three-strain *L. monocytogenes* cocktail at 12 °C for 6 days. PAWs were generated using a plasma bubble reactor using air as working gas (0.8 L/min) at different conditions: 150 V, 1200 Hz, 400 s (PAW A); 150 V, 2000 Hz, 700 s (PAW B); 75 V, 1600 Hz, 550 s (PAW C), and their effectiveness was evaluated after 60 minutes of exposure. Additionally, different coatings were studied, each composed of a base-either (3-Aminopropyl)triethoxysilane (APTES) or N-[3-(trimethoxysilyl)propyl]ethylenediamine (TMSPEA)-and a functional component-either acrylic acid (AcAc) or a solution of poly(ethylene glycol) methyl ether methacrylate (PEGMA). The relative biofilm production was measured by the method of crystal violet and compared with the uncoated SS plates.

Results: Although all coatings assayed decreased the capacity of the pathogenic microorganism to form biofilms, the best result was observed on the surface coated with TMSPEA and AcAc. This combination reached a remarkable 95% reduction compared to the uncoated SS surface. Additionally, all PAWs resulted effective in reducing *L. monocytogenes* biofilms, with reductions ranging from 3.5 (PAW B) to 4.0 (PAW C) log-units after a 60-min treatment.

Conclusion: The current study shows the anti-biofilm activity of plasma-based technologies by preventing bacterial adherence to surfaces and even by inactivating sessile cells, which makes them feasible and cost-effective strategies to be applied in the food industry.

Microbial load in pre-stacked plastic packaging before filling with liquid foods: impact of airborne microorganisms

Pieter-Jan Loveniers¹, Imca Sampers¹, Frank Devlieghere²

¹Research Unit VEG-i-TEC, Department of Food Technology, Safety and Health, Faculty of Bioscience Engineering, Ghent University, Campus Kortrijk, Sint-Martens-Latemlaan 2B, ²Research Unit Food Microbiology and Food Preservation (FMFP), Department of Food Technology, Safety and Health, Ghent University, Coupure Links 653

Aim:

Food packaging plays a pivotal role in maintaining the quality of (semi) liquid food products. While existing regulations focus on the chemical and physical aspects of packaging, microbial aspects, especially in relation to airborne contamination, are often overlooked. This study explores the microbial load on pre-stacked plastic packaging exposed to air in food production facilities, aiming to understand the connection between packaging material and airborne microorganisms.

Method:

Sampling was conducted at nine (semi) liquid food production facilities in Flanders, Belgium. Swab samples were collected from pre-stacked recyclable packaging materials, exposed for 1 hour to the air in the packaging area, during two visits. Airborne microorganisms were sampled using settle plates and impaction techniques. Microbial analysis included Total Microbial Plate Count (TMPC), Yeasts and Moulds (Y&M), anaerobic spores, Lactic Acid Bacteria (LAB), and *Pseudomonas* spp.

Results:

Initial microbial loads on packaging materials varied among companies. TMPC was the most prevalent, detectable in 80% of samples, with a mean concentration of 1.41 ± 0.57 log cfu/100cm² and a range from 0.78 (LOD) to 2.24 log cfu/100cm². Airborne microorganism concentrations ranged widely, with TMPC ranging from 1.00 to 3.49 log cfu/m³. TMPC on packaging materials did not consistently correlate with airborne concentrations, suggesting alternative sources of contamination. Y&M, while detectable in the air, did not always settle on packaging materials.

Conclusion:

The study highlights the potential risk of microbial contamination from pre-stacked plastic packaging materials in (semi) liquid food production facilities. This emphasizes the need for a comprehensive approach to assess microbial dynamics and mitigate contamination risks associated with food packaging materials.

Optimization the disinfection process for flexible packaging using atomized hydrogen peroxide and peracetic acid

Pieter-Jan Loveniers¹, Imca Sampers¹, Frank Devlieghere²

¹Research Unit VEG-i-TEC, Department of Food Technology, Safety and Health, Faculty of Bioscience Engineering, Ghent University, Campus Kortrijk, Sint-Martens-Latemlaan 2B, ²Research Unit Food Microbiology and Food Preservation (FMFP), Department of Food Technology, Safety and Health, Ghent University, Coupure Links 653

Aim:

This study aims to optimize the disinfection process for spouted stand-up pouches using atomized hydrogen peroxide (H₂O₂) and peracetic acid (PAA). The research focuses on integrating atomization technology, which includes fogging and spraying methods, to ensure effective microbial reduction while minimizing residual disinfectant levels.

Method:

The study involved three phases: selecting microbial spores, establishing inoculation techniques, and measuring residual disinfectant levels; assessing drying and disinfection factors such as temperature, airflow, and disinfectant volume; and optimizing drying parameters using a design of experiments approach. Two prototypes—a fogging system and a spray system—were tested for accuracy and effectiveness.

Results:

The fogging prototype initially showed inaccuracies in atomizing hydrogen peroxide, with significant deviations due to leaks and condensation over prolonged periods. Conversely, the spray prototype, equipped with a new pneumatic nozzle, demonstrated improved precision and efficiency. Increasing the drying temperature up to 140°C enhanced drying efficiency and reduced residual disinfectant levels. Bio-validation with *Bacillus atrophaeus* and *Aspergillus niger* confirmed that the optimized parameters achieved substantial microbial reduction.

Conclusion:

The integration of atomization technology, particularly with the improved spray prototype, effectively disinfects spouted stand-up pouches while minimizing residual disinfectant levels. Optimizing parameters such as temperature, spray time, and drying time is crucial for achieving both efficacy and safety. This study provides a viable method for the rapid and effective sterilization of flexible packaging materials, meeting the stringent requirements of aseptic filling and storage

Oil-in-water emulsion and acid concentrations impact inhibition and culturability of a *Bacillus cereus* group strain

Agathe Dutoit¹, Clement Trunet¹, Nicolas Decourcelle¹, Anne-Gabrielle Mathot¹, Pr. Louis Coroller¹

¹Univ Brest, INRAE, Laboratoire Universitaire de Biodiversité et Écologie Microbienne

Aim:

The structure and physico-chemical characteristics of food can influence the physiology of spore-forming bacteria implied in food poisoning and spoilage. The aim of this work is to study the physiology of *Bacillus cereus* in an oil-water emulsion (O/W) supplemented with organic acids.

Method:

A model emulsion was developed consisting of a sonicated mixture (50/50 O/W ratio) of hexadecane-Span80 and nutrient broth-Tween80 added with lactic, caprylic, acetic or lauric acid. This emulsion was inoculated with *Bacillus weihenstephanensis* KBAB4 spores and incubated at 30°C. Cell viability was monitored by flow cytometry over time using Syto9/PI labelling. The cells were enumerated on three agar media: BHI agar (BHA), BHA supplemented with catalase or thiourea.

Results:

Growth inhibition by organic acids was satisfactorily quantified in nutrient broth. However, unexpected cell concentration kinetics were observed in the emulsion. A decrease ($-2 \log(\text{CFU/g})$) followed by a sudden increase ($>5 \log(\text{CFU/g}) / \text{h}$) was observed during enumeration on BHA. Microscopy and flow cytometry analysis confirmed the cell viability and their membrane integrity in the emulsion. The use of catalase or thiourea allows the total bacteria recovery after emulsion exposure. These additives reduce the effect of reactive oxygen species present in BHA, which could inhibit bacterial multiplication. A mathematical model was proposed to describe the maximum proportion of cells unable to grow on BHA, the time to loss of culturability and the time to recovery of culturability.

Conclusion:

These observations highlight viable but non-culturable state of a *B. cereus* group strain in emulsion. The quantification of bacteria in complex matrices is greatly influenced by the food formulation and the enumeration method.

Development of software for evaluation of cold chain disruption duration in foods of animal origin

Doc. MVDr. Ph.D. Lenka Necidova¹, Mgr. Ph.D. Danka Harustiaková², Mgr. Ph.D. Alena Zouharova¹,
Ing. Ph.D. Klara Bartakova¹, Doc. MVDr. Ph.D. Sarka Bursova¹

¹University of Veterinary Sciences Brno, ²Masaryk University

Aim:

During transport and storage, chilled poultry meat products must be kept at a temperature below 4 °C, minced meat below 2 °C, and fish at a temperature approaching that of melting ice (Regulation (EC) No 853/2004). The presented study evaluates the effect of elevated transport temperatures on the outcome of microbiological analyses of selected types of chilled foods of animal origin in simple packaging as well as in vacuum and modified atmosphere packaging. The study aimed to develop software capable of determining the necessity (or absence of the necessity) to perform a microbiological analysis of samples in which the cold chain was disrupted during transport.

Methods:

Model experiments simulated temperature increases during transport of the samples to the temperatures of 4, 8, 11, 14, 17, 20, and 25 °C, with exposure periods of 1, 2, 3, 3.5, and 4 h. Microbiological analyses were performed immediately after the exposure to the elevated temperature (0 h), 3 h, and 24 h after the return to adequate storage temperature. The following microbiological parameters were determined for each sample: the counts of total microorganisms (ISO EN 4833-1/2013), of psychrotrophic microorganisms (ISO 17410/2020), and of *Escherichia coli* (ISO 16649-2/2003). In addition, the presence of *Salmonella* spp. (ISO EN 6579-1/2020) was also assessed.

Results:

The study statistically evaluated the effects of three factors, namely (1) higher temperature, (2) the duration of cold chain disruption, and (3) the examination time after the disruption on the numbers and abundances of microorganisms in samples. The results were used to develop mathematical models describing the effects of temperature and the duration of exposure to the elevated temperature on the microbial profile of food. These results enabled the development of software that quantifies the risk of microbial contamination of foods of animal origin due to cold chain disruption, including risk visualization. The user selects the type of food and its packaging type, the temperature in the transport box (between 5 and 25 °C) to which the sample has been exposed, the duration of exposure to this elevated temperature (from 0.5 to 4 hours), and the length of time the sample is intended to remain stored at a suitable temperature in the refrigerator/import line until microbiological examination (from 0 to 24 hours). The software, which is freely available at <https://webstudio.shinyapps.io/transportvzorku/>, then returns the expected counts of microorganisms and a recommendation for or against the microbiological analysis of the sample.

Conclusion:

These models can serve to establish the maximum acceptable cold chain disruption duration in foods of animal origin. The developed software serves very well (in particular) to the laboratories and supervisory bodies, helping to reduce the number of samples not accepted for processing by laboratories due to improper transport.

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Improving food quality and safety by developing innovative antimicrobial packaging activated with natural bioactive compounds

Valeria Poscente^{1,2}, Claudia Zoani¹, Luciana Di Gregorio¹, Andrea Visca¹, Elisa Clagnan¹, Manuela Costanzo¹, Antonella Del Fiore¹, Benedetto Aracri¹, Luigi Garavaglia³, Francesco Salvadori⁴, Gabriel Mustatea⁵, Nastasia Belc⁵, Roberta Bernini², Annamaria Bevivino¹

¹Department for Sustainability, Biotechnologies and Agroindustry Division, Italian National Agency for New Technologies, Energy and Sustainable Economic Development (ENEA), ²Department of Agriculture and Forest Sciences, University of Tuscia, ³I.L.P.A. Group, ⁴I.L.I.P. s.r.l., ⁵National Research and Development Institute for Food Bioresources - IBA Bucharest

Aim: The prevalence of biofilm-associated microorganisms and the increasing use of ready-to-eat fresh products represent the current duality the food industry must address. Innovative and eco-friendly antibiofilm solutions and appropriate microbiological food control systems are urgently needed to improve food quality and safety. This study aims to develop an antimicrobial active packaging for preserving fresh fruit and vegetables by using combination strategies of natural antimicrobial compounds.

Method: *In vitro* tests were performed comparing culture-based and flow cytometry (FCM) approaches to monitor the cultivability and viability of planktonic and sessile cells after treatments. The best performing conditions were used to develop 5 different antimicrobial packaging evaluated at 7 different sampling times, up to 12 days. Microbiological analyses were conducted to assess total bacteria and fungi counts. Actively growing colonies will be isolate and identify using MALDI-TOF. Migration tests will be conduct for each active packaging condition.

Results: Preliminary results showed the combined efficacy of carvacrol with a pre-formed biofilm monolayer of the probiotic *Lactiplantibacillus plantarum* DSM 20174. The presence of the *L. plantarum* pre-formed biofilm enhanced the carvacrol efficacy, resulting in bactericidal action and significant loss of cell viability, and promoting the use of reduced concentrations. This evidence led to the production of innovative biodegradable and antimicrobial packaging activated with chitosan, nisin as lactic acid bacteria extract (LAB), and carvacrol at different concentrations to extend the shelf-life of fresh "Fayette" strawberries up to 12 days compared to the control.

Conclusion: Our approach integrates advanced and high-throughput microbiological techniques to assess markers of quality and safety, along with chemical analysis of packaging materials. This innovative strategy not only mitigates the environmental impact of the distribution chain but also enhances the value of Italian agricultural products, such as "Fayette" strawberries, promoting sustainability and safeguarding the national agricultural heritage.

The effect of *Linomium algarvense* and *Carpobrotus edulis* extracts as microbial inhibitors on fresh-cut apples

Dr Catarina Pereira², Dr Isabel Ratão^{1,3}, Dr Luísa Custódio², **Dr Célia Quintas**^{1,3}

¹Universidade Do Algarve, ² Centre of Marine Sciences (CCMAR/CIMAR LA), ³MED–Mediterranean Institute for Agriculture, Environment and Development & CHANGE–Global Change and Sustainability Institute

Aim: Halophyte plants are recognized as a source of bioactive compounds (polyphenolics, coumarins, triterpenes, alkaloids), possessing various bioactivities, such as antioxidant, anti-melanogenic, and antimicrobial, among others. The objective of this study was to evaluate the preservation capacity of the halophytes *Linomium algarvense* and *Carpobrotus edulis* extracts on fresh-cut apples.

Method: The ability of the ethanolic extracts of the plants (1 mg/ml and 10 mg/ml) to inhibit microbial growth during 10 days, at 10 °C, was studied by enumerating aerobic mesophilic (AM) (30 °C) and psychrotrophic (AP) (6 °C) microorganisms, and fungi (25 °C) on apple slices using standard methods. Total coliforms and *Escherichia coli* were analysed on Chromocult agar. Control solutions prepared with sterilized distilled water and ascorbic acid (10 mg/mL) were also tested.

Results: *E. coli* was never detected in the apple during the study. Considering the conditions tested (1 mg/ml and 10 mg/ml extracts, sterilized distilled water, and ascorbic acid), in the apple slices treated with *C. edulis* extracts, there was an increase in the microbial populations enumerated over the storage period at 4 °C, as well as with the controls. The AM increased from a range of Log 3.19-Log 4.85 on the first day of the experiment to a range of Log 6.47-Log 7.48 after 10 days of refrigeration. The population of molds varied from Log 2.80-Log 4.54 to values of Log 3.80-Log 4.49 at the end of the assay. No significant differences were found among the results of the four treatments regarding the microbial groups studied, at the end of the storage. On the other hand, the 10 mg/L of *L. algarvense* extract inhibited the growth of the AM (Log 2.96±0.05-Log 2.86±0.76) and AP (Log 3.38±0.09- Log 4.12±0.05) microorganisms assessed as well as the molds (Log 3.77±0.10-Log 2.76±0.21) and yeasts (Log 2.70±0.06-Log 2.78±0.06).

Conclusion: This study shows that extracts of the halophyte *L. algarvense* have the potential to be a source of ingredients with antimicrobial properties useful in apple preservation.

Reduction of allergenic proteins in white mustard seeds

Dr Anna Grygier¹, Dr hab. Dorota Piasecka-Kwiatkowska¹, **Professor Magdalena Rudzińska¹**

¹Poznań University Of Life Sciences

Aim: Every day many people around the world struggle with the problem of allergies. Among several types of allergies, we can distinguish food allergy, which limits the diet plan of people who are allergic to certain food ingredients. One of the food ingredients that may cause allergies is mustard. Mustard allergy is not widespread, but in people who are allergic to mustard it can cause serious effects after consumption. The present study investigated the possibility of reducing allergenic protein levels in white mustard seeds using different types of microorganisms.

Method: In the research were used three different microorganisms for the fermentation of white mustard seeds - *Lactobacillus rhamnosus*, *Saccharomyces cerevisiae*, *Rhizopus oligosporus*. After two days of fermentation, proteins were extracted from the samples. Slot-blot and Western blot analyses were performed to compare the effect of microbial enzymes on allergenic protein levels. Proteins were immobilized on PVDF membrane. The membrane was incubated with the recognition antibody, i.e., human serum from a mustard-sensitized person. Further the membranes were incubated with human IgE labeled with alkaline phosphatase. After washing, the substrate for phosphatase was applied to the membrane. After this step, the reaction was stopped by washing with distilled water. In the next step, electrophoresis of proteins was carried out. Proteins were initially denatured using a solution with SDS. The gradient gel with separated proteins was used to perform Western blot. The separated electrophoretic proteins were transferred with the blotter to a membrane and labeling of individual allergenic proteins for mustard was performed using serum from a human mustard allergic person, human IgE labeled with alkaline phosphatase and a substrate for phosphatase. To stop the reaction, the membrane was washed with water. After the membrane dried, it was possible to analyze it densitometrically using the CLIQS program.

Results: Slot blot analysis resulted in striations on the membrane, which are a response to the presence of allergenic proteins in individual samples. The presence of allergenic proteins was demonstrated in all seed samples. Electrophoresis confirmed the literature information related to the presence of many low-molecular-weight allergenic proteins in mustard. On the other hand, in the Western blot method, results were obtained, confirming the hypothesis of the present project. The presence of allergenic proteins was not demonstrated in the samples of seeds that were fermented by the *Rhizopus oligosporus* strain. Low-molecular-weight (about 10 kDa) allergenic proteins were present in the other samples.

Conclusion: Western blot results do not match those obtained by the Slot blot method. In the case of the Slot blot method, there may have been enough interaction of the compounds present in the samples with the substrate necessary for the analysis. In future studies, it would be appropriate to repeat the Slot blot analysis using a peroxidase-conjugated secondary antibody

Nanoemulsified essential oils reduced tremendously the heat resistance of *Listeria monocytogenes*

Dr. Mariem Somrani¹, Carmen María Pastor¹, Prof. Alfredo Palop¹

¹Universidad Politécnica de Cartagena

Aim:

The implementation of heat treatments stands out as one of the most common preservation technology employed to inactivate pathogens in food. However, food industries are compelled to minimize heat intensity in order to keep the nutritional and sensorial attributes of food and subsequently provide quality products to consumers. To cope with that, the inclusion of essential oils (Eos), being natural, into the heating media has been under investigation to complement heat treatments. EOs have been widely reported as antimicrobials, however in food industry they can pose a challenge due to their immiscibility in water and strong flavor. Therefore, nanoemulsions (NE) offer a solution to overcome these drawbacks..

This study delves into the exploration of the combined effect of heat with nanoemulsions of Lemon (NEL), orange (NEO) and cinnamon (NEC) on *Listeria monocytogenes* inactivation.

Method:

To make the effect of Eos more pronounced and more kinetically stable, they were used as NE during this work. The NE were prepared by ultrasonication. Namely, the aqueous phase was prepared by mixing distilled water and propylene glycol. The oily phase was obtained by adding Tween 80 to EOs and mixed.

Afterward, thermal inactivation kinetics for *L. monocytogenes* CECT 4032 in Tryptone Soy Broth (TSB) supplemented and not with 0.2 ml of NEL, NEO or NEC were determined in a thermoresistometer Mastia at 58°C. Samples were collected in sterile tubes at preset time intervals, diluted and finally incubated.

Results:

The 3 NEs showed an interesting antibacterial effect when added to the heating medium compared to the control. While the NEC reduced the heat resistance by half, NEL and NEO emerged as the stand-out performers, resulting in a reducing the heat resistance of *L. monocytogenes* by about 50 times.

Conclusion:

These findings unveil a very interesting potential of nanoemulsified EOs when combined with heat. On one hand, this approach holds the promise of dramatically reducing the intensity of current thermal treatments applied in the food processing sector. On the other hand, it optimizes the use and integration of essential oils (EOs) into the aqueous phase of food by the means of NE.

Inhibition of pathogenic *Escherichia coli* growth in fruit juice using essential oil emulsions

Prof. Hajime Takahashi¹, Sho Kurokawa¹, Ayaka Nakamura¹, Takashi Kuda¹

¹Tokyo University of Marine Science and Technology

Aim:

The demand for fresh juice that has not been heat-sterilized is increasing as consumers have become more health-conscious. However, consuming unpasteurized apple juice and apple cider can cause pathogenic *Escherichia coli*-mediated food poisoning. Since heat treatment deactivates some nutrients in juice, a method that maintains the nutritional value, while suppressing the growth of contaminant microorganisms, should be developed. In this study, we prepared an essential oil (EO) emulsion and investigated whether it could suppress pathogenic *E. coli* growth in juice.

Method:

First, an EO emulsion was prepared using a high-speed homogenizer and its stability was evaluated by observation and particle size measurements. Sucrose fatty acid ester was used as the emulsifier, and citral and limonene were used as EO. Next, the minimum inhibitory concentration (MIC) of the prepared EO emulsion against *E. coli* O157:H7 was determined. Based on these MIC values, the highly effective citral emulsion was added to apple juice, and its ability to suppress *E. coli* O157:H7 growth was verified.

Results:

The particle sizes of the citral and limonene emulsions prepared were 0.16 and 0.15 μm , respectively, and did not separate for 30 days at room temperature. This revealed that although the particle size differs depending on the type of EO, nanosized emulsions can be created using either type of EO.

The MIC of citral emulsion for *E. coli* O157:H7 was 250–500 ppm, while even 7000 ppm limonene emulsion did not suppress *E. coli* O157:H7 growth. This indicates that citral emulsion is suitable for inhibiting *E. coli* O157:H7 growth. Therefore, we subsequently used only the citral emulsion, which has confirmed antibacterial effects.

E. coli O157:H7 growth in apple juice was significantly inhibited for 7 days after adding 125 ppm citral emulsion. Furthermore, 250 and 500 ppm citral emulsion decreased the *E. coli* O157:H7 count to below the detection limit.

Conclusion:

Citral emulsion can be stored as an additive at room temperature and has a high growth-inhibiting effect on *E. coli* O157:H7 in culture medium and apple juice. The citral emulsion prepared in this study should improve the safety of unpasteurized fruit juices.

Handheld NIR Spectroscopy in Minced Meat Freshness Determination Supported by Machine Learning Algorithms

Dr.rer.nat Isik Riza Türkmen¹, Ervienatasia Djaw¹, Thorsten Tybussek¹

¹Fraunhofer Institute For Process Engineering And Packaging (ivv)

Aim:

This study aimed to leverage machine learning (ML) algorithms for the use of low-cost handheld near-infrared (NIR) spectrometers for a rapid determination of the freshness of beef minced meat preserved under different storage conditions.

Method:

Beef minced meat samples preserved under modified atmospheric packaging (MAP) or under organic conditions were stored at different temperatures and for different time intervals. Reflectance spectra were acquired using a handheld NIR spectrometer SCIO® version 1.1.2. The preprocessing of spectra involved averaging, logarithmic transformation, baseline correction and normalization. Data exploration was done by principal component analysis (PCA). Experimental data were used to train, test and validate a random forest classification model to predict the freshness levels of beef minced meat.

Results:

PCA revealed significant wavelength regions crucial for assessing meat freshness, particularly in the near-infrared regions. The Random Forest model successfully generalized to predict beef minced meat freshness using the provided experimental data collected by using a handheld NIR spectrometer. A prediction accuracy of bigger than 99 % was achieved on both cross-validation and test datasets.

Conclusion:

The robustness of the ML model trained on minced meat preserved under different storage conditions demonstrated a low cost NIR spectrometer's capability to predict freshness without influence from preservation conditions. The promising results suggest the potential for this approach to be applied to other animal and plant-based meat products, supporting rapid and reliable freshness assessment and contributing to food safety and resource sustainability.

Optimization of treatment chamber design for decontamination of almond-based milk alternative by pulsed electric fields

Arisa Thamsuaidee^{1,3}, Matteo Stefanelli², Claudia Siemer¹, Vasilis P. Valdramidis³

¹Elea Technology GmbH, ²University of Parma, ³National and Kapodistrian University of Athens

Aim: To explore the use of pulsed electric fields (PEF) as an alternative to thermal pasteurization of almond-based beverage. Specifically, two different designs of treatment chamber (co-linear and parallel-plate) were compared with regards to the decontamination performance and the contribution of thermal vs. electroporation effects on isolated spoilage microorganisms.

Method: The microbiota of almonds and unprocessed almond beverage was identified via MALDI-TOF MS and/or 16S rRNA sequencing. Separate microbial challenge tests were performed in a continuous PEF system equipped with treatment chamber in either co-linear or the parallel-plate configurations. To differentiate between heat and electroporation, additional work was performed to obtain the thermal inactivation kinetic parameters and the PEF temperature-time profiles for each type of treatment chamber.

Results: *Pediococcus pentosaceus* was the most frequently identified vegetative non-spore forming microorganism in the almond raw materials and unprocessed beverage. In comparison to *Lactiplantibacillus plantarum* WCFS1, *P. pentosaceus* exhibited higher resistance against PEF and thermal inactivation. Nevertheless, as the PEF process intensity increased (as exemplified by the electric field strength and specific energy input), it was possible to achieve $\geq 5 \log_{10}$ reductions of vegetative microorganisms in almond beverage, resulting in a product with extended storage life at refrigeration temperatures. The parallel-plate chamber design was found to have an overall lower temperature impact in comparison to the standard co-linear design.

Conclusion: The current study offers insights into the type of microbial contaminants found in almond-based beverages. While PEF proved to be a promising technology for gentle preservation, it will be crucial to define the target pertinent microorganisms during process optimization, as there could be variability in resistance between different spoilage microorganisms. Furthermore, utilizing a different chamber design could form part of the optimization strategy to overcome the heat sensitivity of valuable food components.

Inactivation of *Listeria innocua* in dry-cured hams by High Pressure Processing and following storage

Food Technologist Elena Dalzini¹, Elena Cosciani-Cunico¹, Muhammad-Ehtesham Abdul¹, Paola Monastero¹, Daniela Merigo¹, Stefania Ducoli¹, Alessandro Norton¹, Marina-Nadia Losio¹

¹Istituto Zooprofilattico Sperimentale Della Lombardia E Dell'emilia Romagna "bruno Ubertini"

Aim:

The industrial implementation of high-pressure-processing (HPP) is constantly increasing due to its efficiency in controlling foodborne pathogens and spoilage microorganisms. HPP has been widely used as an effective post-packaging technology to control *L. monocytogenes* mainly in RTE. Therefore, the purpose of this work was to evaluate the effect of HPP (600 MPa for 5 min) on the inactivation of *L. innocua* (as a surrogate of *L. monocytogenes*) in the interior of whole deboned dry-cured ham with different water activity (*aw*), stored then at 4° and 20 °C for 28 days.

Method:

Three batches of whole deboned hams (with *aw* ranging from 0.82 to 0.93) were sliced and contaminated with a mix of four *L. innocua* strains (ca. 7-8 log CFU/g), vacuum packed, pressurized at 600 MPa for 5 min, and then stored for 28 days at 4° or 20°C. Control samples (non-contaminated) were also treated to check the effect on the physico-chemical properties. Ten samples replicates were analysed for each sampling time (after HPP and at 14 and 28 days during the storage) for the *L. innocua* counts. Three independent experiments were replicated in this study. The log reduction was calculated as $\log(N/N_0)$ where N was the concentration after sampling time and N_0 was that at the beginning of the test.

Results:

In dry-cured hams with $0.82 \leq aw \leq 0.90$ the *L. innocua* counts decreased linearly as a function of the *aw*, with a decrease between 0 to -2 log reduction and then, increasing the *aw* up to 0.92 the log reduction remained unchanged during the HPP. Therefore, during the subsequent storage, a further decrement count was observed from 0 to 2.7 log reduction, depending on the *aw* and on time, with a higher inactivation contributes at 20°C than at 4°C, and at 28 days than 14. No changes in the physico-chemical characteristics were found.

Conclusion:

The pressurized treatment of dry-cured ham with high *aw* and the following vacuum storage, might be additional hurdles to enhance the pathogen inactivation, contributing to the product safety in case of post-processing contamination.

The widely used MicroTester apparatus in the food chain and in the research

Tekla Engelhardt¹, Oktay Hakir, Máté Farkas, Orsolya Strang, Miklós Süth, Ákos Józwiak

¹University of Veterinary Medicine

Aim: Our aim was to show the different applications of the MicroTester. Nowadays there are a lot of detection methods available in the food industry, we just have to decide what our goal is.

Method: The MicroTester is based on the detection of the change in redox-potential caused by microbial activity. The instrument's evaluation system is similar to impedimetric methods, but with a wider range of applications. Redox-potential is one of the most complex indicators of the physiological state of microbial cultures and its measurement is a useful tool for qualitative and quantitative determination of microbial load. The MicroTester was also successfully used to measure the hygiene samples in different environments and to store the data for further analysis.

Results: The MicroTester is a smart tool where we can detect only 1 cell or we can measure the heat resistance for a specific microbe or we can measure hygienic samples within 6 hours (depending on the initial concentration of microbes). The MicroTester is able to perform measurements and reliably determine the cell concentration in a wide range (between 1-10⁷ cells), there is no need for dilution series. The dynamic heat destruction of *Listeria innocua* with and without prior sub-lethal heat treatment has also been quantified using the MicroTester. The MicroTester is able to measure the hygienic status of the environment and the data collected provides the basis for further risk-based analysis.

Conclusion: The MicroTester can be used advantageously for the evaluation of classical membrane filtration, surface swabbing or plate count methods. It is also possible to combine the MicroTester with PCR techniques to perform microbial identification tests. It is also shown that the data collected and stored by the MicroTester can be used as a basis for risk-based analysis in the specific industries where the measurements are made.

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Combining HHP with natural antimicrobials to inactivate *L. monocytogenes* in beef and plant-based patties

Phd Student Nikolaos Giannoulis¹, Mario González-Angulo^{2,3}, Beatriz Melero², Miriam Ortega Heras², Carolina Bocigas Martín², Theocharia Tsagkaropoulou¹, Kimon Andreas Karatzas¹

¹Department of Food & Nutritional Sciences, University of Reading, ²Department of Biotechnology and Food Science, University of Burgos, ³Hiperbaric, S.A.

Aim:

High Hydrostatic Pressure (HHP) is a nonthermal technology widely used in the food industry to extend shelf life and ensure food safety. In the quest to make food processing more sustainable, the combination of HHP with natural antimicrobials emerges as a strategy to use milder processing conditions while meeting food safety objectives. The primary objective of this study was to investigate the potential synergism of HHP in combination with either nisin or a product derived by blueberry extracts to enhance food safety through *Listeria monocytogenes* control in beef and plant-based burgers.

Method:

This study focused on the effect of three processing conditions (300, 400 & 500 MPa for 3 min) in combination with nisin (500 IU/g) or blueberry by-product (4%) on the inactivation of a 5-strain cocktail of *L. monocytogenes* in beef and plant-based burgers. Patties containing the additives were inoculated with 10⁷ CFU/ml of the pathogen cocktail and subjected to HHP. Enumeration was carried out before and after processing in selective agar medium.

Results:

In plant-based burgers, *L. monocytogenes* cells were reduced by 3.8 and 4.0 log units combining HHP (500 MPa) with blueberry by-product and nisin respectively, while a 2.6 log reduction was achieved by HHP alone. Conversely, in beef patties a 4.5 to 5.0 log reduction was observed due to the combined treatments. A synergistic effect of >1 log CFU/g was found in both matrices when HHP (500 MPa) was used with the antimicrobials. In vegan matrix, the synergism was significantly higher when HHP was applied with nisin compared to blueberry by-product. Overall, combining HHP with either nisin or blueberry by-product was consistently more effective against *L. monocytogenes* than HHP alone in both food matrices, irrespective of HHP conditions.

Conclusion:

Based on the observed synergistic effect, this research provides insights into the potential applications of combining HHP with natural antimicrobials as a strategy to enhance the inactivation of *L. monocytogenes*, while optimizing the processing conditions. This aligns with the growing demand for more sustainable and natural systems regarding food production.

High-Pressure Processing Inactivation for Foodborne Bacteria, Viruses and Parasites in Foods

Kah Yen Claire Yeak¹, Cristina Serra-Castelló¹, **George Pampoukis**¹, Sara Bover-Cid², Berta Torrents-Masoliver², Heidy M.W. den Besten¹, Marcel H. Zwietering¹

¹Food Microbiology, Wageningen University, ² Food Safety and Functionality Program, IRTA

Aim:

High-pressure processing (HPP) is a non-thermal technology used for microbial inactivation of food products. In this study, 22 microbial hazards (i.e. pathogenic bacteria, parasites, and viruses) were classified for their resistance to HPP. Also, the effective pressure and time combinations were estimated and the main factors of impact were identified.

Method:

The data from 248 articles were extracted and the inactivation kinetics were converted to the logarithm of the decimal reduction time; $\log_{10}D$ (\log_{10} min) ($n=3890$) and analyzed in three ways. Firstly, the general inactivation trend was investigated for different microbial hazards and pressure levels. Secondly, an exploratory data analysis identified potential predictors through correlations, statistics, and linear models. Additionally, it ranked the microbial hazards at three reference pressure levels: 400, 500, and 600 MPa. Lastly, the potential predictors were used for multiple linear regression and mixed effects models.

Results:

Five groups of microorganisms were formed, namely, i., sensitive (*Toxoplasma*, *Cyclospora*, *Cryptosporidium*, *Trichinella*, Rotavirus, *Aeromonas*, *Cronobacter*, *Campylobacter*): $D_{400MPa} < 1$ min; ii., moderately resistant (Norovirus, Hepatitis A, *Listeria*, *Salmonella*, *Yersinia*): $1 \text{ min} \leq D_{400MPa} < 3$ min; iii., resistant (Hepatitis E, vegetative *B. cereus*, *S. aureus*, *E. coli*): $3 \text{ min} \leq D_{400MPa} < 5$ min; iv., highly resistant (*Mycobacterium*, *Shigella*): $D_{400MPa} \geq 5$ min; and v., spores (*B. cereus*, *C. botulinum*, *C. perfringens*): no inactivation at 400 MPa. The inactivation of bacterial spores was due to thermal effects. For non-sporeforming microorganisms, the main factors of impact were pressure, pH, and the a_w of the treated matrix. The model developed using pressure, food item, and the microbial hazard as fixed effects terms, and the strain as a random effect term had conditional $R^2=0.7$, showing that the inactivation was species and strain-dependent.

Conclusion:

All three analysis methods identified similar classifications of microbial hazards. Viruses and *Enterobacteriaceae* showed varying resistance depending on their genus. With only a few key parameters, namely, pressure, microbial hazard, and the food item, a large amount of the observed variance could be explained. The microbial classification created in this study details various pressure and time combinations to reduce microbial hazards, which can guide regulatory authorities and industrial applications.

Combining non-thermal plasma technology with hygienic design for food and water safety for fresh-cut lettuce

Dr. Uta Schnabel¹, Dr. Clemens Morath², Jörg Stachowiak¹, Andreas Ell³, Prof. Dr. Oliver Schlüter⁴, Dr. Jörg Ehlbeck

¹Leibniz Institute For Plasma Science And Technology, ²GARTENFRISCH Jung GmbH, ³KRONEN GmbH,

⁴Leibniz Institute for Agricultural Engineering and Bioeconomy

Aim: Bacteria, including human pathogens and zoonoses, are known for their (persistent) biofilm growth. This can be a particular challenge in the production of fresh/fresh-cut food, as high humidity and nutrient availability provide a perfect basis for biofilm growth. Equipment with hygienic design, antimicrobial properties of the process water and CIP/SIP processes offer the opportunity to minimise/avoid biofilm growth. Here, plasma-processed-air-treated water (PTW) was combined with hygienic design in a commercial lettuce washing line to investigate its industrial applicability and antimicrobial efficacy.

Method: PTW was used in a lettuce washing line to investigate the total viable count (TVC), microbial viability and vitality of the fresh-cut lettuce itself and the process water with/without storage, taking into account food quality and toxicology.

Results: Depending on the PTW concentration, TVC reductions of 2 lg steps in lettuce and 3 lg steps in process water were detected. Viability and vitality were also reduced. Food quality such as colour was not affected and no toxicological effects were observed. The use of PTW in different washing steps was realised under industrial conditions.

Conclusion: The promising results of this project, the strong industrial need and the realistic implementation possibilities offer new non-thermal plasma-based tools for the challenging control of unwanted biofilm growth in the food industry.

Enzymatic grafting of phenolic compounds to pectin as a pathway for functional ingredients development

Pamela Freire de Moura Pereira¹, Amparo Jiménez-Quero¹

¹Chalmers University Of Technology

Aim:

Dietary fibers have received great importance in function of their beneficial effects on the gastrointestinal tract, and therefore to the human health.[1] In plant cell wall, these dietary fibers can be found natively attached to phenolic compounds which can burst the bioactivity of such macromolecules.[2] Mimicking this pattern, it is possible to modify the carbohydrates structure by the coupling of phenolic compounds, imparting bioactivity and modifying its physico-chemical properties.[3,4] The present study is focusing on the modification of pectin molecules, by an enzymatic coupling of phenolic compounds (ferulic and caffeic acid, as well as free phenolic compounds from coffee byproducts products), mediated by the usage of laccases. The modified polysaccharide will be further applied as active edible packaging materials.

Method:

Coupling validation is assessed through FTIR and UV-vis. The bioactivity of the modified pectin is correlated in terms of concentration of phenolic compounds and antioxidant activity of the polysaccharide. The effect of pectin modification on its thermal properties is assessed by thermogravimetry (TGA) and differential scanning calorimetry (DSC).

Results:

The enzymatic coupling of phenolic compounds to pectin structure enhanced the antioxidant activity of the compounds, contributing as well for modifications on their physico-chemical properties.

Conclusion:

The modification of pectin with phenolic compounds represents a promising path for development of functional ingredients, with potential applications on active edible packaging applications.

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In vitro Synergistic Activity of Natural Antimicrobials Combinations for Potential Antimicrobial Active Packaging Application

Mr Paul Gichuki Kamau¹, M.C Cruz-Romero¹, PA Calderon¹, J.P Kerry¹, M.A Morris²

¹Food Packaging and Materials Science Group, School of Food & Nutritional Sciences, University College Cork, ²AMBER and the School of Chemistry, Trinity College Dublin

Aim: To assess the antimicrobial activity (AA) of 6 natural antimicrobial (NAM) combinations against pure cultures of pathogenic and spoilage bacteria (*Escherichia coli*, *Listeria innocua*, *Pseudomonas fluorescens*, *Brochothrix thermosphacta*, *Staphylococcus aureus*, and Lactic acid bacteria) and fish microflora for potential application in the development of Antimicrobial Active Packaging (AAP) materials.

Method: To determine the synergistic (Fractional Inhibitory Concentration Index (FIC) ≤ 0.5) effect of 6 NAM combinations of low molecular weight chitosan (CS) and essential oil (EOs) (CS: Origanum EO (OEO), CS: Thyme EO (TEO), CS: Garlic EO (GEO), TEO: OEO, TEO: GEO and OEO: GEO) the checkerboard assay was used. To confirm the effectiveness of the synergistic activity of the NAM combinations, time-kill test was performed.

For the attachment of the NAM combinations onto vacuum skin packaging (VSP) films, NAM combinations (4X) were added to Gelatine (8% w/v) solution containing glycerol (30% w/w) and coated onto VSP films. Quantitative (ASTM E2180) and qualitative (disk-like agar diffusion) tests were carried out to determine the AA of the coated VSP films.

Results: From the NAM tested, individually CS exhibited the highest AA against all bacteria tested including fish microflora with minimum inhibitory concentration (MIC) of 0.125 mg/mL. The EOs (TEO, GEO and OEO) exhibited a MIC value of 0.313 mg/mL for all bacteria except for *Pseudomonas fluorescens* and fish microflora which were more resistant (MIC of 0.625 mg/mL). The synergistic activity nature of the 6 NAM combinations indicated that the concentration of each NAM used in the combination decreased significantly ($P < 0.05$) when compared to their individual MICs indicating that less of the NAM in question is required in the combination to deliver the same antimicrobial effect. The time-kill test indicated that the NAM combinations were bactericidal. Quantitative and qualitative AA tests indicated that the developed AAP materials had a good AA against all bacterial culture tested.

Conclusion: Overall, results indicated that synergistic NAM combinations have the potential to enhance AA and that the developed AAP materials can potentially be used as antimicrobial packaging for shelf-life extension of food products thus reducing food waste and enhance sustainability.

Influence of drying on the sea fennel extract properties used for active fish packaging

Iva Čanak¹, PhD Mario Ščetar¹, Auree Jacob², Phd Marko Nuskol¹, PhD Kata Galić¹

¹University of Zagreb, Faculty Of Food Technology And Biotechnology, ²IUT Dijon, University of Burgundy

Aim:

Given the recent surge in scientific interest towards isolating phenolic compounds from natural sources, *Crithmum maritimum* L. (sea fennel), a self-sustaining halophyte native to the Mediterranean basin, emerges as a cost-effective and valuable resource for the food and packaging industries. This study aims to investigate the impact of drying methods (air drying, lyophilization, and infrared drying) preceding microwave-assisted extraction, as a green and sustainable technology, on the properties of water and hydroalcoholic sea fennel extracts intended for incorporation into edible fish packaging.

Method:

Fresh *Crithmum maritimum* L. leaves were harvested, washed with tap water and spin dried. Then, three drying methods were applied to prepare the plant for extraction: air drying (23±2°C, 10 days), lyophilization (24 hours), and infrared drying (15 minutes at 110°C). The plants were homogenized using a blender and stored in vacuum-sealed bags at 4°C in the dark. Extracts were obtained through microwave-assisted extraction (Milestone Start S, 500W, 80°C, 10 minutes), with varying plant/solvent ratios and solvent types (50% (V/V) ethanol and water). Total phenolics were quantified spectrophotometrically, and antioxidant potential was assessed through two in vitro assays: ferric reducing antioxidant power (FRAP) and 2,2-diphenyl-1-picryl-hydrazyl (DPPH). Fresh extracts were utilized to formulate chitosan-based film-forming solutions, with their pH, viscosity properties, and miscibility evaluated.

Results:

The maximum extraction of phenolic compounds was achieved with 50% ethanol, with air-dried samples exhibiting the highest values (49 mg gallic acid equivalent/g plant) and infrared-dried samples the lowest (8.9 mg GA/g plant). FRAP values ranged from 60 to 725 mg ascorbic acid equivalent/g, and DPPH inhibition ranged from 37 to 84%. Water extracts of sea fennel proved to be suitable candidates for chitosan-based formulations, demonstrating favourable properties without adversely affecting miscibility, pH, or viscosity.

Conclusion:

This study pioneers the evaluation of drying pretreatments combined with sustainable extraction methods for sea fennel. It underscores the potential of sea fennel extracts in chitosan-based coatings, particularly beneficial for oxidation-sensitive foods like fresh fish. Optimal pretreatment conditions are crucial for extracting valuable bioactive compounds from underused Mediterranean plants, emphasizing their significance in food and packaging industries.

Development of active packaging from agar/carboxymethyl cellulose bioplastic with integrated Green Silver nanoparticles

Dr Seyedeh Fatemeh Mirpoor¹, Dr Alessio Massironi², Dr Stella Lignou¹, Dr Sameer Khalil Ghawi¹, Dr Dimitris Charalampopoulos¹, Federico Trotta²

¹Department of Food and Nutritional Sciences, University of Reading, P.O. Box 226, Whiteknights, Reading RG6 6AP, UK, ² Metalchemy Limited, 71-75 Shelton Street, London WC2H 9JQ, UK

Aim:

Replacing synthetic plastics in food packaging with environmental friendly materials developed from renewable and biodegradable sources can effectively address the significant contribution of plastics to environmental pollution. Therefore, producing bioplastics from natural resources such as polysaccharide, which are non-toxic, biodegradable and relatively cheap, is highly desirable. In addition, the incorporation of silver nanoparticles (AgNPs) into the bioplastic can improve the shelf life of the packed food, as it has been shown to have an antimicrobial effect against pathogenic microorganisms.

Method:

The bioplastic was produced by casting a blend of agar and carboxymethylcellulose, and different concentrations of silver nanoparticles (AgNPs) synthesised using environmentally friendly methods were incorporated into the film matrix to develop an active packaging. Several physicochemical properties of the bioplastic were investigated such as antimicrobial activity, water sensitivity, mechanical and thermal properties.

Results:

The obtained results revealed that the addition of a higher concentration of AgNPs resulted in an increase of the tensile strength and young's modulus of the film, which indicates a more resistant and rigid material obtained after the incorporation of AgNPs in the matrix. Conversely, the elongation at break of the film prepared in the presence of a higher concentration of AgNPs decreased slightly. The thermogram of the AgNP-loaded composite film showed similar behaviour to that of the neat agar/cellulose film, indicating that the thermal stability of the film was not affected by the addition of AgNPs. The antimicrobial activity test of the films against *E. coli*, *Listeria monocytogenes*, *Staphylococcus aureus* and *Pseudomonas aeruginosa* revealed that the control film had no antimicrobial activity against the tested microorganisms whereas the film containing 1.5 w/w% AgNPs showed antimicrobial activities against all the tested bacteria. The water sensitivity of the films decreased when the higher concentration of AgNPs was incorporated into the film, however the moisture content was not affected.

Conclusion:

The developed bioplastics in the presence of higher concentration of AgNPs becomes stiffer and less sensitive to water with antimicrobial activity against pathogenic microorganisms. This bioplastic can be a suitable alternative to conventional plastics with the ability to extend the shelf-life of food products packed in it.

Enhancing Sustainability in Food Packaging: Synthesis and Characterization of CMC/NIPU Bioplastics with SLS Incorporation

Student Nazila Oladzadabbasabadi¹, Prof Benu Adhikari¹, Distinguish Prof Elena Ivanova¹, Doctor Mehran Ghasemlou^{1,2}

¹School of Science, STEM College, RMIT University,, ²Centre for Sustainable Bioproducts, Deakin University

Aim

Sustainability has become a crucial element in food packaging, with the industry constantly striving to integrate sustainable practices into its products. Petrochemical polyurethanes (PUs) are extensively used in many food packaging containers due to their outstanding properties, including resistance to abrasion, thermoplastic behavior, durability, and toughness. However, the synthesis of these PUs relies on using isocyanate precursors, which are typically considered toxic. The notable drawbacks associated with isocyanates, such as high toxicity and phosgene-based synthesis, have prompted substantial efforts aimed at developing environmentally friendly processes or sustainable alternatives. In this study, we advance the field by synthesizing pre-synthesized NIPU and hybrid materials using carboxymethyl cellulose (CMC)/NIPU, with the incorporation of sodium lignosulfonate (SLS) to enhance sustainability and performance.

Method

Non-isocyanate polyurethanes (NIPUs) can be formed from the reaction between cyclic carbonates and polyamines. The corresponding NIPU products are environmentally safe and not harmful to humans. We present the synthesis of NIPUs through a green and facile solvent-free and catalyst-free reaction between cyclic carbonates (propylene carbonate) and diamines (1,2-ethylenediamine). We will then comprehensively elucidate the development and full characterization of food packaging bioplastics made of CMC and NIPU incorporating SLS.

Results

The evaluation of mechanical, thermal, and structural analyses demonstrated judicious interactions between CMC and NIPU at the level of 20 wt%. Leveraging this optimized level, we will endow additional strength to resultant bioplastics by utilizing their synergistic interactions with lignin. Furthermore, the hybrid material exhibited superior UV-blocking, antioxidant, and antimicrobial activities.

Conclusion

We successfully synthesized non-toxic NIPU via a solvent-free and catalyst-free route. The incorporation of 20% NIPU into CMC has demonstrated high flexibility and elasticity. The hypothesis regarding the inclusion of SLS suggests that it not only enhances the mechanical strength of our bioplastics but also imparts additional functionalities such as antioxidant and antimicrobial properties. Food packaging materials containing SLS may also exhibit UV-blocking capabilities, which is particularly beneficial for UV-sensitive food products. These bioplastics, with their enhanced functionalities, are anticipated to find widespread applications across various industries, especially in food packaging.

Influence of pH on the acrylamide content of canned black olives

Doctor Concepción Romero Barranco¹, Mercedes Brenes Álvarez¹, Doctor Eduardo Medina Pradas¹, Doctor Manuel Brenes Balbuena¹, Doctor Eva María Ramírez Castro¹, Doctor Pedro García García¹
¹Instituto de la Grasa (IG), Spanish National Research Council (CSIC), Ctra. Utrera km 1, Building 46, 41013

Aim: Black ripe olives are considered a “low-acid canned food” and must be sterilized in order to be safe. Acrylamide, a potential carcinogen, is formed in black olives during the thermal treatment. It is known that the formation of acrylamide is dependent on the pH of the olive flesh. The aim of this study was to evaluate the addition of acids and calcium chloride in the packing brine on the acrylamide formation and quality parameters (color and firmness) of the final product.

Method: Darkened olives of the Hojiblanca cultivar were bottled in A314 jars. Variables were the followings: (i) fruit presentation (whole or pitted); (ii) pH before packaging; and (iii) addition of acids (lactic/citric) and calcium chloride (6g/L). After sterilization (121°C, F₀=15), jars were stored at room temperature for 2 months, and analyzed their acrylamide content along with quality parameters (surface colour and texture).

Results: Commercial presentation was significant for the content of acrylamide because pitted olives contained lower concentration of acrylamide than whole olives. Moreover, the pH of the olive flesh before heating was a determinant parameter for acrylamide formation. Sterilised fruits with a pH value close to neutrality possessed higher concentration of acrylamide than those with an alkaline pH value. The addition of acids in the brine of fruits with an alkaline pH made them more palatable along with a the decrease in acrylamide concentration that could reach up to 20% in whole olives and about 10% in pitted olives. Likewise, the addition of calcium chloride allowed a more optimal firmness in the final product.

Conclusion: A reduction of up to 20% in acrylamide was achieved packing the black olives with slightly alkaline pH. The addition of acids and calcium chloride did not affect acrylamide formation although it increased the quality of the final product.

Characterization of Gelatin-Based Centrifugally Spun Active Fibers Containing Bay Laurel Leaf Essential Oil

Gunes Su Guler¹, Sila Basturk¹, Dr. Nalan Yazicioglu², **Professor Servet Gulum Sumnu**¹, Professor Serpil Sahin¹

¹Department of Food Engineering, Middle East Technical University, ²Department of Nutrition and Dietetics, University of Health Sciences

Aim

Active food packages aim to to maintain the food quality and safety throughout the shelf-life of foods. Bay leaves, which has the origin name as *Laurus nobilis* L. (Lauraceae), is a plant contains essential oil and used in foods, drugs and cosmetics. It has various benefits such as antioxidant, anti-inflammatory, anticancer and antiviral activity. Gelatin, a renewable and biodegradable polymer, is a promising external support material to encapsulate bay leaf essential oil. Therefore, the objective of the study is to develop gelatin based active fibers with bay leaf oil by using centrifugal spinning which is an efficient and a novel method to produce fibers.

Method

In this study, bovine gelatin was used as a polymer with 20% (w/v) concentration, by dissolving it overnight in 100% acetic acid. Then, 1, 3 and 5% (v/v) bay leaf oil and 0.2 grams of Tween80 were added separately and stirred for 30 minutes. For the centrifugal spinning, the volumetric flow rate and the rotating spindle speed were set as 20 mL/h and 1800 rpm, respectively. Viscosity of solution, diameter, antioxidant activity and total phenolic content of fibers were determined to examine the effect of bay leaf oil concentration on to the centrifugal spin solution and fiber characteristics.

Results

Viscosity of solutions were 0.341, 0.376, 0,421 and 0.441 Pa.s for control (without bay leaf oil), 1, 3 and 5% bay leaf oil concentrations, respectively. Diameter of the fibers increased as bay leaf oil concentration increased from 1541.4 nm to 2478 nm for control fibers and 5% bay leaf oil concentrations, respectively. Similarly, as bay leaf oil concentration increased, antioxidant activity and total phenolic content of fibers increased. Antioxidant activities were 10.89%, 30.64% and 43.96%, while total phenolic contents were 0.88, 1.58 and 1.74 mg GAE/g for 1%, 3% and 5% bay leaf oil concentrations, respectively.

Conclusion

As concentration of bay leaf oil increased, viscosity of fiber forming solutions and diameter of fibers increased. Addition and increasing bay leaf oil resulted in higher antioxidant activity and total phenolic content. Therefore, bay leaf oil added gelatin fibers might be a promising active packaging material to preserve foods.

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Monitoring microbial growth under well-controlled gas conditions: towards more sustainable food packaging

Msc Seren Oguz¹, Eleonora Bonanni¹, Carlos Marquez¹, Vincenzo Turrisi¹, Lotta Kuuliala¹, Mariem Somrani^{1,2}, Frank Devlieghere¹

¹Ghent University, ²Universidad Politécnica de Cartagena

Aim:

The food packaging industry is continuously seeking more sustainable alternatives to conventional packaging systems. Within the context of modified atmosphere packaging (MAP), this transition requires assessing the impact of the new configuration on the evolution of the atmosphere and, consequently, on microbial growth. However, despite the importance of this knowledge in packaging technology development, the exact relations between different gases and microbial growth are still poorly understood. Addressing this major knowledge gap calls for extensive data collection utilizing research infrastructures that allow precise controlling of gas composition over storage time. Hence, this study aimed at establishing guidelines for monitoring microbial growth in liquid media under controlled atmospheres.

Method:

The fundamental requirements (most importantly, factors to be non-controlled, controlled, or eliminated) were identified through the literature review. The technical requirements were determined based on a dataset representing *Listeria monocytogenes* growth in liquid brain heart infusion (neutral and acidified) at two temperatures (4 °C and 7 °C) under well-controlled atmospheres (CO₂%/O₂%/N₂%; 0/0/100, 20/0/80, 40/0/60, 60/0/40, 60/2/38, 60/20/20, 0/20/80).

Results:

Decisions on which variables to eliminate or control were made to streamline the experimental process for determining the fundamental relations between the gas(es) and growth. Intrinsic factors (e.g. food chemical composition) and implicit factors (e.g. background flora) should be eliminated or controlled because their interactions with the target culture are unpredictable. Extrinsic factors such as gases are mostly interconnected and should be controlled during storage to understand their impact on microbial growth. Therefore, an airtight Gas-washing Bottle Incubation System (GBIS) was utilized to precisely control atmosphere, pH, and temperature, equilibrate the growth medium with the desired atmosphere, and facilitate non-destructive sampling. Standardized media enabled accurate measurement and reliable quantification of *L. monocytogenes* growth. This study discerned individual effects, including reduced growth with higher CO₂, lower pH, and lower temperature.

Conclusion:

The data acquisition process in a liquid medium elucidates quantitative interactions between many variables and microbial growth under static conditions. This methodology, facilitated by GBIS, can be extended to food safety of liquid food, fermentation, and chemistry. The insights gained from studying *L. monocytogenes* growth contribute to designing specific sustainable packaging solutions.

Brewers' spent grain: development of edible coating for strawberry preservation

Ph.D. Student Ilary Belardi¹, Solange I. Mussatto², Assunta Marrocchi³, Ombretta Marconi¹

¹Department of Agricultural, Food and Environmental Sciences, University of Perugia, Via San Costanzo, 06121 Perugia, Italy, ²Department of Biotechnology and Biomedicine, Technical University of Denmark, Søtofts Plads, Building 223, 2800 Kongens Lyngby, Denmark, ³Department of Chemistry, Biology and Biotechnology, University of Perugia, Via Elce di Sotto, 8, 06123 Perugia, Italy

Aim:

This study repurposes brewers' spent grains (BSGs) to create sustainable edible coatings, addressing concerns about plastic packaging. BSGs, consisting in high moisture, proteins, and lignocellulose, represent a key brewing by-product. The potential for BSGs to provide proteins, phenolic compounds and oligosaccharides by enzymatic and hydrothermal extraction enables the formulation of edible coatings to improve the strawberries' shelf-life. This work aims to promote eco-friendly practices in both brewing and packaging industries, aligning with the circular economy approach.

Method:

BSGs from Carlsberg were processed at DTU Bioengineering, Technical University of Denmark. Using sequential enzymatic (pH 8.5, 100 µL/g of protease) and hydrothermal (150°C, 60 min) treatments, proteins, phenolic compounds, and oligosaccharides were extracted from wet BSGs. A starch-based solution (2% w/v starch in deionized water) was formulated, with varying concentrations (10%, 2%, 3% w/w) of proteins, phenolic compounds, oligosaccharides, or their combinations. Coated and dried strawberries were then stored for 12 days, and their quality assessed for decay percentage, weight loss, pH, and DPPH scavenging activity.

Results:

Enzymatic hydrolysis yielded the highest protein extraction yield (65%) and phenolic compounds concentration (566 mg GAE/g dry matter). It also resulted in a 20% extraction yield of oligosaccharides, consisting of gluco-, xylo-, and arabinoxylo-oligosaccharides in a ratio of 29:44:27 (w/w/w). Upon evaluating the coated strawberries after 3 days of storage, the results shown less mouldy coated samples (11-30%) with unaltered pH (3.5) than uncoated control (44-54% and 2.7, respectively). Coated strawberries with both compounds had lowest weight loss (3%) compared to the uncoated control (9%). Further, the DPPH scavenging activity ranged 33.1-41.9 % of inhibition (day 3) compared to the 12 day (4.2-7.9%).

Conclusion:

BSGs represent a rich source of bioactive compounds with various applications. Our study found that edible coating enriched with proteins, phenolic compounds, and oligosaccharides significantly improved preservation and quality of strawberries compared to control samples. These findings suggest that the formulated edible coatings have potential for extending the shelf-life and maintaining strawberries' quality. Further research to optimize coating formulation and application methods could enhance their benefits for food preservation, contributing to sustainability and food security efforts.

Exploring the Potential of Pineapple Waste Parts in Agar Film Production: Characterization and Performance Analysis

Nivin Varghese¹, Carlota Costa¹, Bruno Marques¹, Diogo Pereira¹, Dr. Clara Sousa¹, Dr. Adma Melo¹, Dr. Inês Ramos¹, Prof. Cristina LM Silva¹, **Dr. Teresa Brandão¹**

¹Universidade Católica Portuguesa, CBQF—Centro de Biotecnologia e Química Fina—Laboratório Associado

Aim:

Agar films have gained attention as biodegradable packaging materials due to their sustainability and versatility. This study explores the potential use of pineapple waste parts (rind, core, and crown) as a renewable source for agar film production. The aim is to investigate the characteristics and performance of agar films incorporating powdered pineapple subproducts for enhanced packaging applications.

Method:

Control agar films were prepared by boiling and stirring a mixture of water, agar (2% w/v), and glycerol (10% w/w of the agar). Pineapple waste parts, including rind, core, and crown, were freeze-dried, ground, sieved (particles with diameter < 250 µm were used), and then incorporated into the film forming solutions by substituting 25% of the agar weight. Each sample film was obtained by pouring 20 g of the solutions into petri dishes (diameter of 9 cm) and dried at 35 °C for approximately 20 hours. The films were kept under ambient conditions and characterized in terms of color (Lab* coordinates), thickness, pH, water activity, moisture content, mechanical properties (tensile strength and elongation at break), and contact angle (for wettability assessment). Additionally, Fourier Transform Infrared Spectroscopy (FTIR) analyses were conducted for further characterization and films comparison.

Results:

When pineapple waste was added to the formulations, color differences increased, especially noticeable in those with crown powder, resulting in darker, more opaque films. Thickness was consistent across all films (0.134 ± 0.096 mm), except for crown-derived ones, significantly thicker (0.322 ± 0.198 mm). Films were equivalent in terms of water activity (0.515 ± 0.013) and moisture content ($14.86 \pm 0.66\%$). However, there was variation in pH; agar-control (6.75 ± 0.01) and crown-based films (5.71 ± 0.02) exhibited significantly higher pH values compared to the others (3.73 ± 0.03). Films with pineapple subproducts showed reduced tensile strength and elongation at break compared to the control. Contact angle differences were not significant, suggesting similar wetting behavior. FTIR analysis indicated varied compositions, which may justify the mechanical performance differences.

Conclusion:

Agar films enriched with pineapple subproducts exhibit promising characteristics for potential packaging applications. However, further exploration into their water vapor and oxygen barrier properties is warranted.

Enhancing Biodegradable Film Properties: Protein-Chitosan Conjugation for Improved Water Vapor Barrier and Durability

Dr. Sevgin Dıblan^{1,2}, Prof. Dr. Zafer Erbay²

¹Food Processing Department, Vocational School of Technical Sciences at Mersin Tarsus Organized Industrial Zone, Tarsus University, ²Department of Food Engineering, Faculty of Engineering, Adana Alparslan Turkes Science and Technology University

Aim: Growing concerns regarding petroleum-based plastic waste driving global ecological pollution have prompted researchers to pursue new, durable, and biodegradable materials with superior film properties. Biodegradable films, particularly edible ones for food packaging, have been extensively researched but often lack crucial attributes such as water vapor barrier and durability. Recent efforts have made strides in improving the mechanical properties of edible films made from protein-carbohydrate combinations. Nevertheless, these films remain structurally unstable with insufficient water vapor barriers. This study aims to develop more durable films by utilizing a biopolymer solution (whey protein isolate or sodium caseinate) combined with chitosan (WPI-CTS and NaC-CTS) through a Maillard-type reaction.

Method: The objective is to enhance water vapor barrier properties and introduce functional attributes to the films by leveraging protein conjugation for enhanced performance. The Maillard-type conjugation occurred naturally under specific reaction conditions. An optimization approach was applied to achieve the optimal WPI-CTS and NaC-CTS biocomposite, focusing on parameters such as pH (3-5), temperature (60-80°C), protein: chitosan ratio (10:1-10:5), and time (8-24 hours). The optimized sample underwent further characterization analyses including FTIR, XRD, and TGA.

Results: Subsequently, films were produced under optimized conditions, and evaluations were conducted on mechanical strength and water vapor permeability to assess the impact of conjugation on film properties. The results indicated that conjugation between chitosan and WPI or NaC led to improved films compared to non-conjugated ones.

Conclusion: With these results, the potential for enhancing film properties through conjugation becomes apparent. Therefore, in future studies, it can be aimed to utilize these conjugated biopolymers as wall materials in emulsion films to improve water permeability, thus addressing a primary drawback of such food packaging materials. This research was supported by The Scientific and Technological Research Council of Turkey (project no: 122C074).

Towards sustainable food preservation: harnessing bioactive molecules from vegetable sources for functional eco-friendly bio-coatings

Andrea Cavallero¹, Laryssa Peres Fabbri¹, Francesca Vidotto¹, Laura Aliotta^{2,3}, Vito Gigante^{2,3}, Dr. Cristiana Sbrana¹, Dr. Laura Pucci¹, **Dr. Morena Gabriele¹**

¹National Research Council, Institute Of Agricultural Biology And Biotechnology, ²Department of Civil and Industrial Engineering, University of Pisa, ³National Interuniversity Consortium of Materials Science and Technology (INSTM)

Aim: Nowadays, the demand for fresh fruits and vegetables is significantly increasing, and preservation of food post-harvest horticulture crops represents a challenging issue, which looks at cutting by 2030 half per capita the food waste worldwide at both the retail and consumer level. In this context, using extracts from plants and food-waste biomasses offers attractive options to achieve proper coating formulations that can reduce the burden from fossil resources and promote a biobased economy. The project PRIN 2022 - EXTRAFRESCO (Prot. 2022LSTY2Y) aims to extract bioactive molecules from vegetable and low-valorized agro-waste sources (microalgae, hemp seeds, and wheat bran) to obtain a functionalized bio-coating useful in the food industry. The bioactive molecules extracted will be fine-tuned by coupling with appropriate additives, plasticizers, primers, and dispersing agents to develop water-based and solid coating solutions.

Methods: To optimize the extraction procedure and the yield/activity of each extract, different methods (homogenization, UAE-ultrasonic-assisted extraction, and MAE-microwave-assisted extraction) and percentages of water-ethanol solutions (water, 50-100% ethanol, v/v) were used, and the phytochemical content (polyphenols, flavonoids) and antioxidant activity (FRAP, ORAC) of extracts were analyzed at T0 and T14 (14-days after extraction).

Results: Preliminarily, extraction of wheat bran with 50% ethanol achieved better results in terms of polyphenols than water ($p < 0.01$) and 100% ethanol ($p < 0.001$) with the highest content obtained using MAE, followed by UAE and homogenization. Similarly, the most effective extraction of flavonoids was achieved with both water ($p < 0.001$) and 50% ethanol ($p < 0.001$) compared to 100% ethanol. Regarding antioxidant activities, no differences were observed among different extraction methods using water and 100% ethanol, while better results were obtained using 50% ethanol and MAE (5 min 1000W 70°C). Bioactive content and antioxidant properties were stable along with 2 weeks at 4°C.

Conclusion: Preliminary results suggested that MAE could be more suitable for phenolic component extraction from wheat bran extracts and better antioxidant activities. Further conditions and parameters will be analyzed to optimize and select the best extractive procedure to reduce cost, process time, energy, raw material, and environmental impact. We acknowledge financial support under the National Recovery and Resilience Plan (NRRP), Mission 4, Component 2, Investment 1.1, Call for tender No. 104 published on 2.2.2022 by the Italian Ministry of University and Research (MUR), funded by the European Union – NextGenerationEU – Project Title EXTRAFRESCO – CUP I53D23002210006.

Characteristics of biobased film-forming solutions made from i-carrageenan and enriched with green and brown algae

Mia Kurek¹, **Iva Čanak**¹, PhD Mario Ščetar¹, PhD Kata Galić¹

¹Faculty of Food Technology and Biotechnology

Aim:

Efforts to develop edible films for extending the shelf life of sensitive foods, such as fish, have been ongoing. This study focused on optimizing formulations using iota-carrageenan, a polysaccharide derived from red seaweed, known for its unique film properties. Several formulations were studied using rheometric techniques and the content of i-carrageenan, plasticizers and green/brown microalgae was investigated as alternative fish packaging compared to traditionally used materials.

Method:

Film-forming solutions comprised iota-carrageenan (1,5 to 3 % m/V) and plasticizers (glycerol and sorbitol, 0 to 30 % of polymer dry weight). To achieve better gelation, film forming solutions were heated to 70 °C (for 20 min), cooled to 25 °C, poured on appropriate casting surfaces and dried overnight in a ventilated climatic chamber. Films were enriched with green (*Spirulina* spp.) and brown (*Ascophyllum nodosum*) microalgae at different concentration (0, 1 and 5 % m/m). The rheological characteristics and pH of starting film-forming solutions, were evaluated. The physico-chemical and functional properties of edible films were determined by the content of base material, plasticizer, and microalgae.

Results:

Initial solutions were basic (pH 9-9.5), with pH decreasing as iota-carrageenan and plasticizer concentrations increased. Microalgae addition further decreased pH. Viscosity increased with temperature due to gelatinization. Samples exhibited non-Newtonian behavior, with consequently expected flow index higher at 70°C (0.22-0.67) than at 23°C (0.61-0.72). Plasticizer addition increased flow index while decreasing k value (consistency index), indicating enhanced molecular entanglement. Microalgae addition impacted flow behavior and casting properties. Optimization involved adjusting polymer concentration, plasticizer content, and microalgae type and amount. Increasing glycerol percentage didn't affect gelatinization temperature but reduced final viscosity, yielding less viscous films.

Conclusion:

The study optimized biopolymer film formulations for fish packaging by adjusting polymer concentration, plasticizer type, and microalgae content. Increasing glycerol percentage didn't affect gelatinization temperature but reduced final viscosity, yielding less viscous films. These findings contribute to the development of sustainable packaging solutions for application of coatings for fish preservation.

Microwave-assisted ethanolic and aqueous extraction of antioxidant compounds from tomato and lemon by-products

Lorena Martínez Zamora^{1,2}, Laleh Mozafari¹, Rosa Zapata¹, Marina Cano-Lamadrid¹, Prof. Encarna Aguayo¹, Fuensanta Melendreras³, Jenaro Garre³, Francisco Artés-Hernández¹

¹Universidad Politécnica de Cartagena, ²University of Murcia, ³National Technology Centre for Food and Canned Food -CTNC-

Aim: A total of 124.3 Mt of citrus were produced in 2023. Around 34% is destined for juice production, where up to 80% of inedible waste is generated. On the other hand, annual tomato production round about 177.1 Mt and processing generates a waste of 37%, approximately.

The aim of the present work was to study the effect of a microwave-assisted ethanolic and aqueous extraction in the recovery of antioxidant compounds from tomato and lemon by-products.

Method: Ethanolic solvent ratio was 100% EtOH, 50:50 EtOH:H₂O, and 100% H₂O. The amount of TPC was assessed by the Folin-Ciocalteu method and TAC by ABTS and DPPH free radical uptake spectrophotometric methods of the extracts obtained from the MWAEs.

Results: The TPC content in lemon peel by-products was twice that obtained in tomato by-products. It was observed that in the case of tomato a better yield was obtained at 100% EtOH and 50:50 EtOH:H₂O. The extraction yield in lemon peels was the opposite, decreasing as the EtOH concentration increased. Maximum values of 9.4 g GAE kg⁻¹ were obtained in tomato and 19 g GAE kg⁻¹ in lemon peels. TAC by ABTS method were 2 g TE kg⁻¹ higher than those obtained by DPPH, reporting maximum values of 3.56 g TE kg⁻¹ and 6.11 g TE kg⁻¹ in tomato and lemon, respectively.

Conclusion: Aqueous MWAE in lemon and 50:50 ethanolic:water MWAE in tomato could be considered as a green technology to extract a large amount of polyphenols and other bioactive compounds from citrus and tomato discards with great interest for the food or pharmaceutical industry, putting in value agri-food by-products in a circular economy.

Comparative physico-chemical characterization of organic flours from wheat, rye and spelt

Doctor Sérgio Sousa¹, Doctor Pedro Castro¹, Doctor Joana Costa¹, Doctor Francisca Casanova Bastos¹, Dr. Ema Dias², **Prof. Ana Gomes¹**

¹Universidade Católica Portuguesa, CBQF - Centro de Biotecnologia e Química Fina – Laboratório Associado, Escola Superior de Biotecnologia, Rua Diogo Botelho 1327, 4169-005, ²Germen - Moagem de Cereais S.A. Rua Joaquim Pinto, 91 4460-339

Aim:

There is an increasing consumer demand for healthier food products, such as cereal flours, free from pesticides and other contaminants. The production of organic cereals is therefore a solution that can be commercially explored. Moreover, consumers also highly appreciate and value the local production of cereals and flours.

Hence, the aim of the current work was to characterize organic flours obtained from three distinct cereals, namely wheat, rye and spelt. The cereals were selected on the basis that they are able to be grown in Portugal as it presents favourable climatic conditions.

Methods:

Physico-chemical characteristics of each organic cereal flour, crucial for adequate processing, were analysed through distinct methodologies/technologies, such as falling number (enzyme activity), mastersizer (size distribution) and scanning electron microscopy (SEM; morphology), among several others.

Results:

Results showed that, among the three flours, wheat presented the highest falling number (350), which translates the enzymatic activity of α -amylase (being negatively correlated). Hence, the dough prepared with wheat flour will be harder than those obtained with the flours from the other cereals. Concerning size distribution of the flours' granules, results showed bimodal distribution regarding wheat and rye flours, while spelt flour resembled a normal distribution. A maximum (peak) was observed for all flours between 20-30 μm , and a second peak was registered at 60 and 135 μm for wheat and rye, respectively. Those results, obtained utilizing a mastersizer, were further corroborated *via* SEM analysis. X-ray diffraction analysis provided information concerning crystallographic structure, which allowed to determine that the flour with lower cristallinity was rye. Since this characteristic decreases susceptibility to enzymes, it is inferred that rye flour will be the easiest to digest. Differential scanning calorimetry (DSC) showed similar starch gelatinization temperature between the three flours, although in spelt flour the process was initialized at lower temperature. Fourier-transform infrared spectroscopy (FTIR) analysis revealed similar spectra, disclosing, as expected, the presence of starch and gluten.

Conclusion:

Knowledge on the physico-chemical properties of each particular organic cereal flour is crucial and must be considered when looking to select one to develop a specific food product.

Screening of mediterranean agro-food by-product as biocomposite fillers for sustainable footwear production

Doctor Tânia Ribeiro¹, **Isa Silva**¹, Doctor Sara Silva¹, Professor Manuela Pintado¹

¹Universidade Católica Portuguesa, CBQF - Centro De Biotecnologia E Química Fina – Laboratório Associado, Escola Superior de Biotecnologia

Aim:

Plastic materials contribute 4.5% to global greenhouse gas (GHG) emissions. The footwear industry contributes significantly to these emissions due to the high plastic use (38-71%) in shoe components. With the aim of reducing the footwear industry's environmental footprint, this study investigated the potential to produce and assess the potential of agro-food by-products (olive pomace (OP), grape stem (GP), vine shoots (VS)) as biocomposite fillers for footwear components. After milling these abundant Mediterranean agro-waste raw materials, physical and chemical composition analyses were performed to evaluate their potential as biocomposite fillers.

Method:

Particle size (sieves: 250,150, 10, and 50 μm), proximate composition (moisture, ash, protein, fat, and carbohydrates), and phenolic content were evaluated. Followed by extractives, cellulose, hemicellulose, and lignin content determination, according to Sluiter et al. 2008 and Sluiter et al. 2021 respectively. Four fractions were also produced and evaluated regarding their extractives, cellulose, hemicellulose, and lignin composition. FTIR analysis was also attained for the fractions with and without extractives.

Results:

After milling, a higher fraction of particles above 250 μm (56%) was attained to OP, and VS had fewer particles above 250 μm (37%). VS displayed the highest carbohydrate content (87.2%), followed by GS (77.3%) and OP (67.5%). VS also exhibited the highest cellulose content (39.7%) and the lowest extractive amount (16.1%). Instead, OP P showed a relatively high composition of extractives (38.2%) and lignin (20.5%). GS has a similar cellulose content (26.4%), lignin (30.6%), and extractives (30.3%) but reveals the highest phenolic content, followed by OP and VS. OP appeared as the most promising reinforcement filler due to its higher yield of smaller particle size fractions, which facilitated better interaction with polymers. However, VS had a more favourable lignocellulosic composition, with high cellulose, low extractives, and lignin content.

Conclusion:

Overall, the investigated agro-food by-products hold promise as valuable sources of lignocellulosic fillers for biocomposite production in the footwear industry. Nevertheless, optimisation of filler preparation processes is essential. While VS and GP require enhanced particle size reduction, OP and GP would benefit from removing or reducing extractives, lignin, and hemicellulose to ensure optimal performance.

Exploring Grain Silo Residues as Lignocellulosic biomass source for sustainable footwear biomaterials

Isa Silva¹, Doctor Tânia Ribeiro¹, Doctor Sara Silva¹, Professor Manuela Pintado¹

¹Universidade Católica Portuguesa, Cbqf - Centro de Biotecnologia e Química Fina – Laboratório Associado, Escola Superior de Biotecnologia

Aim:

Over the years, replacing plastic-based materials has been crucial in alleviating the environmental impact across industries. The footwear industry currently relies on plastic-based materials with a considerable environmental footprint. Lignocellulosic matrices are suitable filling agents due to their structural properties. Hence, sustainable sources rich in lignocellulosic content, such as cereal byproducts, present a promising avenue for novel biomaterials. On a large scale, cereals are stored silos when awaiting transportation, which, when cleaned, accumulate grain silo residues composed of variable cereals and dust. This study aims to explore grain silo powder residues as potential sources of lignocellulosic matrices for footwear manufacturing, considering their composition and particle size variations across different batches.

Method:

Particle size (sieves: 250,150, 10, and 50 μm), proximate composition (moisture, ash, protein, fat, and carbohydrates), and phenolic content were evaluated in three silo residue batches (A, B, and C). Followed by extractives, cellulose, hemicellulose, and lignin content determination, according to Sluiter et al. 2008 and Sluiter et al. 2021 respectively. FTIR, total phenolic content and antioxidant activity (ABTS) were evaluated. Particle size fractions of the three silo residue batches were also characterised.

Results:

Regarding the particle size, batches A and C predominantly consisted of particles in the 100-50 μm range and < 50 μm , whereas batch B mainly contained particles in the 100-150 μm and 100-50 μm ranges. The total phenolic content was higher in batch B, followed by A and C. Consequently, batch C showed better antioxidant activity. An increase in total extractives was also observed when decreasing particle size. The lignocellulosic components were more present in batch B, followed by batch C and A. The FTIR analysis showed similar spectra between batches with some differences in carbohydrate bands.

Conclusion:

In conclusion, grain silo powder residue can be a source of lignocellulosic matrices, but composition and particle size variations can affect their application. Before their incorporation as fillers in bio-based shoe production, it is essential to implement a standardisation procedure of composition, identify the raw materials with the best characteristics for this end, and use the defined parameters as a quality criterion.

European seabass and Gilthead seabream: impact of alternative feed on fat quality indexes

Ana Vulić¹, Tina Lešić¹, Nina Kudumija¹, Jelka Pleadin¹, Dražen Oraić¹, Ivana Giovanna Zupičić¹, Snježana Zrnčić¹

¹Croatian Veterinary Institute

Aim: Aquaculture products are an important source of food for consumers worldwide and aquaculture production is constantly increasing. Although aquaculture production has a lower carbon footprint compared to livestock farming, the constant growth in this sector brings with it some negative impacts, including a high demand for fish feed. The aim of this study was to evaluate the fatty acid profile and lipid quality indices of two fish species important for aquaculture, European seabass and gilthead sea bream, when fed diets containing insects and algae as alternative protein sources.

Method: The chemical composition of the fish was determined by analyzing chemical parameters: water, total protein, fat and ash using standard and in-house analytical methods. The extracted fat was used for the preparation of fatty acid methyl esters prior to GC-FID analysis. Based on the fatty acid profile, the lipid quality indices atherogenicity index (AI), thrombogenicity index (TI) and the ratio of hypocholesterolaemic and hypercholesterolaemic fatty acids (HH) were calculated.

Results: The high content of polyunsaturated fatty acids, i.e. eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), the favorable ratio between omega-3 and omega-6 fatty acids and the good fat quality indices make fish a desirable component of the human diet. The predominant fatty acid was oleic acid, with no statistically significant difference between the groups fed different diets. The results showed that the fat quality indices of both fish species in both commercial and experimental diets were in line with the recommended values. The AI and TI for Gilthead sea bream and European seabass were below 1, the ratio of omega 6 to omega 3 was below 4 and the ratio of PUFA to SFA was above 0.44. A statistically significant difference ($p > 0.05$) was only observed in the HH index when comparing different feed and it was observed for both fish species.

Conclusion: The results suggested that insects and algae can be used as an alternative protein source in fish feed without affecting the fat quality of the fish. Optimization of the fish feed formula is necessary to improve the content of hypocholesterolaemic fatty acids and thus the HH index.

Intelligent poultry feed formulations to improve the sustainability of meat production

Ir. Daphne Michels¹, Dr. Sarah Verkempinck¹, Karen Vermeulen², Riet Spaepen², Professor Emily Burton³, Dr. Dawn Scholey³, Dr. Alexandra Wealleans², Professor Tara Grauwet¹

¹Ku Leuven, ²Kemin Europe N.V., ³Nottingham Trent University

Aim:

Poultry meat consumption rises yearly while other meat types show decreasing popularity. This popularity is largely due to chicken's nutritional composition possessing all essential amino acids, higher protein digestibility than crops, and lower lipid content than other meats. However, its production entails a substantial environmental impact which needs to be reduced to sustainably meet consumers demands. One strategy to reach this objective is by lowering the feed conversion ratio (FCR), i.e. the amount of feed needed to produce a quantity of meat. Emulsifiers and oil are often added to feed to improve lipolysis in bile salt and lipase deficient young broilers, but also starch and protein digestion improve as this approach increases transit times. An undiscovered potential to further improve macronutrient digestion however lies in the emulsification of oil and emulsifiers before addition to the feed instead of relying on their emulsification in the gastrointestinal tract (GIT).

Method:

Broiler diets were supplemented with (i) oil and an emulsifier mix (LEX) separately or (ii) with an o/w emulsion. Different surfactant-to-oil ratios were considered as well: 0.03 and 0.20. Body weight and lipid and protein digestibility were measured *in vivo*. Identical diets were subjected to *in vitro* digestion simulating the growing broiler GIT to gain additional mechanistic insights into macronutrient digestion kinetics.

Results:

FCR's of o/w emulsion supplemented diets (ii) significantly improved compared to diets (i) with values ranging between 1.34-1.40 instead of 1.59-1.64 over the course of three weeks. The largest win was made in the first week with FCR's decreasing from 2.83-2.88 in diets (i) to 1.56-1.67 in diets (ii). However, *in vivo* digestibility trials did not show significantly higher lipid digestibilities of O/W diets. Complementary *in vitro* lipolysis however showed a sigmoidal behaviour with shorter lag times in both diets containing more LEX and/or diets (ii) compared to diets (i). Additionally, different *in vitro* digestion kinetics of lipolysis and proteolysis of diets lacking LEX indicated interactions between lipids and protein in these diets.

Conclusion:

This study showed that emulsion supplementation affects macronutrient digestion kinetics and improves FCR, therefore offering a possible solution in improving poultry meat production sustainability.

Comparative sustainability assessment of alternative protein sources for meat substitution

Prof. Sergiy Smetana^{1,2}

¹DIL German Institute of Food Technologies (DIL e.V.), ²Institute of Food Quality and Food Safety, University of Veterinary Medicine Hannover, Foundation

Aim

The modern food system is characterized with high environmental impact, which is in many cases associated with increased rates of animal production and overconsumption. The adoption of alternatives to meat proteins (insects, plants, mycoprotein, microalgae, cultured meat, etc.) might potentially influence the environmental impact and human health but could also trigger indirect impacts with higher consumption rates. Current study provides a condensed analysis on potential environmental impacts and unintended trade-offs (i.e., nutrient content decrease) associated with integration of alternative proteins in meat substitutes.

Method

The analysis was conducted using the Google Scholar database based on review of original studies published in scientific journals in English during the last decade (till 2022). Studies were selected by applying the keywords “meat” and “protein” plus “substitute”, “analog”. Such a search yielded around 3800 articles. Further inclusion of terms such as “LCA” or “life cycle assessment” or “environmental impact” or “carbon footprint” further limited the number of studies to 81, further narrowed via analysis to 64 sources.

Results

Plant-based meat substitutes on average had a considerably lower greenhouse gas emissions (GHGE) than animal products: farmed fish (34% lower); poultry meat (43%), pig meat (63%), farmed crustaceans (72%), beef from dairy herds (87%), and beef from beef herds (93%). Plant-based extrudates (intermediate products) had low GHGE: 7.7-7.9 kg CO₂eq. kg⁻¹ which was in lower range compared to chicken meat protein 7.7-11.3 kg CO₂eq. kg⁻¹. They were significantly lower in GHGE (2-22.35 kg CO₂eq. kg⁻¹ protein) than hypothetical cultured meat (average 56 kg CO₂eq kg⁻¹ protein). Similarly, a few-fold improvement potential was observed in several categories (terrestrial eutrophication, acidification, photochemical oxidant formation, particulate matter, ozone depletion) for plant fiber products compared to chicken meat.

Conclusion

Studies successfully reflect on indirect environmental, economic, and social factors, as well as resource and environmental impact trade-offs. A further model, based on interaction between the elements of complex food systems and able to define the second and third order impacts (e.g., rebound effects), would be required to predict the influence and role of meat substitutes in future diets and potential shifts with the inclusion of other protein alternatives.

Enhancing Sustainability Through Apple Pomace In Gluten-Free Bread: Nutritional Approach To Life Cycle Assessment

Leire Cantero¹, Estitxu Villamor², Olaia Martinez^{1,3,4}, Ortzi Akizu-Gardoki⁵, Silvia Matias¹, Maite Pérez-Azurmendi¹, Jonatan Miranda^{1,3,4}, Jesús Salmerón^{1,3,4}, Jone Guenetxea-Gorostiza^{6,7}, Jon Esparta¹

¹GLUTEN3S Research Group, Department of Farmacy and Food Science, University of the Basque Country (UPV/EHU), ²EKOPOL Research Group, Department of Physics (UPV/EHU), ³Gluten Analysis Laboratory, Department of Nutrition and Food Science (UPV/EHU), ⁴Bioaraba Health Research Institute, Nutrition and Food Safety Research Group, ⁵Life Cycle Thinking Group, Department of of Graphic Design and Engineering Projects (UPV/EHU), ⁶Department of Preventive Medicine And Public Health, University of the Basque Country (UPV/EHU) , ⁷Biogipuzkoa Health Research Institute, Mental Health Group

Aim

The increasing need and demand for sustainable products has led to strategies in the agri-food industry to mitigate environmental impact, including waste valorization. Market-available gluten-free bread (GFB) lacks nutrition, sensory, and technological aspects compared to its gluten-containing counterparts. Thus, our proposal involves incorporating apple pomace powder (APP) to achieve a high-quality and sustainable GFB. It is a high fiber ingredient (77.8%) derived from the cider industry, obtained by washing, dehydration, and grinding. The aim of this study was to analyze the environmental impact reduction associated with the utilization of APP integrating a nutritional focus and comparing renewable and conventional energy sources.

Methods

To assess the environmental impact, Life Cycle Assessment (LCA) was applied following a cradle-to-grave approach. Three GFB formulations with varying quantities of APP (0%, 6%, 10%) were analyzed, along with two waste treatment processes: conventional and renewable energy mixes. OpenLCA2.1.1 software, databases (Ecoinvent3.9.1, Agribalyse3.0.1) and experimental data were utilized. Two functional units (FU) were employed: a daily portion of bread (100g) , at laboratory scale, and its fiber content. Results were calculated with the ReCiPe-midpoint (hierarchist) method, and global warming potential (as kgCO₂ eq.) was analysed.

Results

With 100g of bread as the FU, when waste treatment was conducted using a conventional energy mix, there was an increase in global warming, primarily due to dehydration stage conditions (0.83% growth when increasing APP from 0 to 10%). However, with renewable electricity, the impact decreased as the amount of APP increased (9.76% reduction from 0 to 10% APP). On the other hand, as the fiber contribution becomes the FU, a reduction in global warming was observed with higher quantities of APP, regardless of the energy source type used in the waste treatment (from 0.027 kg CO₂ eq (0% APP) to 0.017 kg CO₂ eq (conventional mix, 10%APP) and 0.015kg CO₂ eq (renewable mix, 10%APP).

Conclusion

Reusing food waste enhances sustainability and aligns with circular economy principles, with renewable energies emerging as a viable alternative for waste valorization. Nutritional LCA represents an innovative approach; by integrating human nutrition and environmental health within food systems, improvements could contribute to both areas.

Hyperspectral imaging for prediction of evolving physicochemical soil properties in cleaning processes

Mr. Matthew Moore¹, Prof Peter Fryer¹, Dr Ian McRobbie², Dr Nick Dixon², Prof Zhenyu Zhang¹

¹University Of Birmingham, ²Innospec Ltd.

Aim: Understand food cleaning processes for reducing water and energy usage

Improving cleaning technologies to reduce water and energy usage in the food sector would lead to a significantly enhanced sustainability and minimal environmental impact. However, industrial standard methods for assessing the performance of cleaning technologies do not provide an objective and quantitative evaluation, nor insights into the temporal evolution of soil properties during the cleaning process. We are developing a non-invasive methodology to investigate the cleaning process from a physicochemical perspective in real time, using hyperspectral imaging.

Method: Hyperspectral imaging and multivariate analysis in a custom-built cleaning rig

A cleaning rig was developed in which a continuous flow of cleaning solution was passed over a surface food foulant, and hyperspectral imaging in the short-wave-infrared (SWIR) range (1100-1900 nm) was performed at intervals throughout the cleaning process. Image analysis techniques were used to evaluate the total soiled area over the course of cleaning, which allowed “cleaning profiles” for each trial to be obtained. Multivariate analysis was used to predict physicochemical properties of different food soils over the course of various cleaning procedures.

Results: Model and prediction of food soil properties using hyperspectral data

SWIR hyperspectral imaging of the surface foulant at different stages of cleaning showed notable spectral changes which were linked to the evolving physicochemical properties using multivariate analysis. The evolving soil properties were dependent on the method of soil preparation and the method of cleaning employed. “Cleaning profiles” elucidated which conditions improved cleaning performance.

Conclusion: Improved methodology for assessing cleaning performance

The developed methodology represents a significant improvement over industrial standard methods for a more rigorous assessment of cleaning performance and gives insight into the kinetics and interfacial phenomena that govern hard surface cleaning processes. This knowledge can be leveraged to develop sustainable solutions for reduced water and energy usage in cleaning processes.

Integrative bioprocess design for sustainable cultured meat production

Chantal Treinen¹, Marie-Luise Schlieker¹, Katharina Brenner¹, Laurenz Köhne¹, Marius Henkel¹

¹Cellular Agriculture, TUM School of Life Sciences, Technical University of Munich

Aim:

One way towards sustainable human nutrition is to research novel food resources and to strive for efficient food production. To achieve this for cultured meat, several hurdles must be overcome. From a technical point of view, this includes suitable bioreactor systems, process upscaling and costs of culture media, to name but a few. Therefore, one research focus is on 'bioprocess design for cultured meat' with the objective to provide integrative technologies and infrastructure to achieve sustainable cultured meat production.

Method:

The applied methods can be categorized as follows: (i) evaluation of scaffold materials in terms of biocompatibility, physico-mechanical properties etc., (ii) design and characterization of perfusion bioreactor systems, (iii) optimal media utilization and recirculation, (iv) process monitoring and intelligent process control assisted by mathematic modeling, (v) process simulation as a basis for techno-economic analysis (TEA) and life cycle assessment (LCA).

Results:

A scale-down approach is used to investigate scaffold materials and perfusion bioreactor systems regarding their suitability for cultured meat production. To this end, prototypes of perfusion chambers were designed using CAD (computer-aided design) and manufactured with FDM (fused deposition modeling) 3D printing. Additional progress has been made in evaluating various scaffolding materials and shapes, applying e.g. gellan gum, alginate, collagen or gelatine. To ensure efficient utilization of medium components, such as cost-intensive growth factors, the incorporation of a recycling unit is anticipated. Real-time process monitoring remains a challenge, which is why another key aspect is the development of intelligent process control, e.g. through mathematical modeling or the use of soft sensors. A process model of the cultured meat production is established using SuperPro Designer, also considering downstream processing and waste streams. Based on the simulation, the production process can be analyzed via TEA and LCA to identify environmental hotspots.

Conclusion:

To ensure efficient and environmentally friendly cultured meat production, integrative bioprocess design is required. The interdisciplinary nature of bioprocess engineering allows the production process to be viewed from multiple angles. The simulation of the overall production process and the associated iterative optimization help in pursuing realistic concepts and to create new solutions for a sustainable nutrition.

Food waste management in Lebanese hospitals

Professor Mohamad Abiad¹, Hussein Hassan, Maha Hoteit

¹American University Of Beirut

Aims

This study sought to assess the food waste management protocols in different types of food services at Lebanese hospitals.

Methods

This observational, descriptive, cross-sectional study sampled 32 dietitians in Lebanese Hospitals using a convenience sampling technique, who were interviewed to complete a questionnaire.

Frequencies and percentages were obtained through simple statistics.

Results

Study findings showed that, in Lebanese hospitals, 81.3% of food waste was obtained from patient's tray, 66.7% from expired prepackaged food, 58.1% from hospital cafeterias, and 48.4% from expired-unconsumed cafeteria food. The food waste obtained is mixed with general waste. None incorporates composting or anaerobic digestion due to financial issues, but 18 hospitals are open to considering these methods. About 85% of hospitals adopt different approaches for managing food waste from isolated patients. Only 18.8% had external contractors for waste management, among which 9.4% mentioned food waste management in the contract.

Conclusion

Lebanese hospitals rely on garbage disposal as a food waste management technique, lacking effective strategies, such as composting or anaerobic digestion. This underscores the urgent need for comprehensive and improved food waste management measures in the country. This study is a wake-up call for policymakers to adopt a robust approach in formulating hospital food waste management policies.

Beyond the Tray: Analyzing Hospital Food Waste for Economic and Environmental Sustainability

Professor Mohamad Abiad¹, Hussein Hassan, Maha Hoteit

¹American University Of Beirut

Introduction: Reducing and managing food waste offers several benefits for people and the environment, such as increased food security, climate change mitigation, cost savings, and less strain on the land, water, biodiversity, and waste management systems. Hospital food services generate significant food waste daily; thus, hospital waste has gained much attention in previous years.

Aim: This study aims to determine the magnitude of food waste in Lebanese hospitals and to highlight its economic and environmental repercussions.

Methods: A descriptive cross-sectional study was conducted between April 2023 and September 2023 and involved 155 inpatients from cardiovascular, gastrointestinal, obstetrics-gynecology, and surgical wards of 16 hospitals across the Lebanese governorates.

Results: The plate waste represented 31.4% of the amount served, equating to an annual waste volume of 366 tons in the participating hospitals. The mean waste cost was USD 1.04 per hospital bed per day. On average, waste from a hospital bed resulted in the emission of 1.57 kg CO₂-eq, the loss of 457 liters of fresh water, and 3.17 g of nitrogen to the environment.

Conclusion: Lebanese hospitals should take pivotal steps to reduce and manage food waste and eventually alleviate its adverse impacts on the country's fragile economic system and scarce environmental resources.

A food waste framing can help promote purchase of suboptimal potatoes

Dr Jeanine Ammann¹, Dr Carole Liechti¹, Dr Gabriele Mack¹, Dr Rita Saleh¹

¹Agroscope

Aim

Discarding visually suboptimal vegetables is a huge problem in terms of food waste. We therefore aimed to test whether food waste information framing would make suboptimal potatoes more attractive to consumers.

Methods

Data were collected in 2024 through an online survey conducted in Switzerland using a mixed methods approach. The final sample consisted of 481 participants (51% women, mean age: 47 years). Our 2 × 2 (setting × information) experimental design randomly assigned participants to one of four experimental conditions. For the setting, we chose farm shops versus supermarkets, and for the information (treatment), we provided food waste information (explaining that potato B is commonly sorted out) or none (control). In each condition, participants were asked to choose between perfect-looking potato A, as is commonly available, or suboptimal potato B, which is commonly sorted out, or neither. Potatoes A and B were equally expensive to control for price. Subsequently, the participants explained why they had made this choice.

Results

Our setting comparison indicated that the participants showed a higher tendency to choose potato A (optimal looking) in the farm shop setting than in the supermarket setting. However, adding food waste information increased the probability of participants choosing potato B (suboptimal). The groups differed significantly ($\chi^2(3, n = 469) = 14.71, p < 0.01$), indicating that the information provided had a significant effect on the potato choice. Across all conditions, price was a strong reason for choosing potato A. Some participants indicated that potato B should be cheaper than potato A. Others explained that they would cut out the bad spots from potato B, which would result in a better price–performance ratio for potato A. The reasons for choosing potato B were distinctly different. In the food waste condition, participants most often indicated that potato B was still suitable for consumption or that they chose it to reduce food waste.

Conclusion

Our study showed that providing food waste information can motivate consumers to buy suboptimal potatoes and thereby help reduce food waste. These findings are promising and should be translated into practice.

Quantification of waste in fruits and vegetables using computer vision-based image analysis

Assist. Prof. Burce Atac Mogol¹, Ezgi Dogan Comert¹, Vural Gokmen¹

¹Hacettepe University

Food loss and waste (FLW) remains a major problem globally, estimating one-third of all food produced is lost or wasted. According to the UNEP (2021) report, 17% of total global food production is wasted. Among them household waste comprise the highest portion (61%) especially with the waste in fruits and vegetables, as perishable foods. In order to effectively manage and reduce FLW, it is essential to accurately quantify the amount of FLW.

Aim:

The main reason for the waste of fruits and vegetables at household level is typically attributed to their decay, which also leads to changes in color. Color changes during processing or storage of fruits and vegetables can be monitored by image analysis techniques. Computer vision-based image analysis is a non-destructive method for extracting information from images of objects. This study aims to determine the amount of waste in fruits and vegetables using computer vision-based image analysis.

Method:

In this study, this technique was employed to quantify the rotten portions of fruits, serving as a waste measurement tool at the household level. Initially, images of individual fruits and vegetables were captured from various angles (top and side views). The rotten sections of the fruit were then identified and isolated by image segmentation using an algorithm developed in MATLAB. The percentage of the rotten part, calculated from image, was correlated with the weight of the wasted part of the fruit. The relation was modelled by means of kinetics approach.

Results:

The results showed that the formation kinetics of rotten part fits well to the first order kinetics and the estimation could be achieved until certain apparent maximum. The apparent maximums were found as 38%, 26%, 39.6%, 56.2% for apple, pear, lettuce, and orange, respectively. The kinetic behaviors of decay were found to be specific to fruits and vegetables.

Conclusion:

This study showed that actual waste (weight %) of certain fruits and vegetables could be quantified just from their image by using image analysis.

Shelf life prediction of beverage bases with multivariate accelerated shelf life testing

Linda Katsch, Arthur Gossen¹, Martyna Bator¹, Jan Schneider¹

¹OWL University of Applied Sciences and Arts

Aim:

Reducing food losses and waste is an important goal of today's sustainability efforts. The aim is to develop a dynamic shelf life for the storage and transportation of beverage bases for lemonade. If the quality can be monitored during this period and the reactions taking place are known, it may be possible to create a dynamic shelf life. In the future, this model should be used in a monitoring device that accompanies the beverage base along the value chain and can send current data.

Method:

The quality of the beverage bases used for the production of raspberry and orange lemonade was evaluated using a range of chemical and physical parameters. The samples were stored under accelerated storage conditions at temperatures of 20, 30, 40, 50 and 60 °C. The data were analyzed using the multivariate accelerated shelf life testing (MASLT) method. The various parameters were summarized in principal component analysis (PCA) and the multivariate kinetic parameters were calculated from this.

Results:

The results demonstrated that temperature- and time-dependent changes in various quality parameters could be detected in the beverage base. These changes could be evaluated using PCA and the MASLT algorithm could be applied. A time law could be applied, and the activation energy and acceleration factors could be calculated. From this, the shelf life during storage in the temperature range from 20 to 60 °C can be calculated using a cut-off point.

Conclusion:

The MASLT method enables the combination of various individual quality parameters into a single value, the calculation of kinetic parameters, and the creation of a model to predict a dynamic shelf life thereby reducing waste in the beverage industry.

A systematic literature review on impactful food waste interventions at consumer level.

Dr. Carole Liechti¹, Dr. Gabriele Mack¹, Dr. Jeanine Ammann¹

¹Research Group Economic Modelling and Policy Analysis, Agroscope

Aim:

Until today, in dept-knowledge about impactful food waste interventions at consumer level is lacking. This review aims thus to assess the impact of the intervention to reduce food waste at consumer level.

Method:

We conducted a systematic literature review in September 2023 according to the PRISMA protocol. A total of 49 publications with interventions aiming reducing food waste were selected and assessed based on three defined evaluation schemes from the literature. These were: six intervention categories (including single and multi-component interventions), the robustness of the study design (with low, medium and high) and the impact of the interventions on food waste reduction (with none, low [non-significant reduction] and high [significant reduction]).

Results:

Results showed that the major part of publications examined predominantly single interventions (77.5%), while multi-component interventions (22.5%) were less utilized. Among the single-component interventions, nudging (54%) and training and knowledge enhancement strategies (27%) were most represented. Overall, 65.5% of the food waste interventions assessed in our review exhibited a medium level of robustness in their study design. Study designs with a high robustness accounted for 24.5%, while those with low robustness comprised 10%. Results showed that most of the interventions had a high impact- (71.5%) while less than a third had a low impact (22.5%) on food waste reduction. Few interventions were classified as having none or a negative impact (6%) on food waste reduction. Further, all multi-component interventions showed a high impact on food waste reduction, while only 62% of all single interventions were classified with a high impact. However, the reported maximum levels of food waste reduction between multi-component and single interventions were similar: nudge with training and knowledge enhancement intervention with up to 79% ($p=0.001$) and nudge intervention with up to 78.2% ($p<0.001$).

Conclusion:

Multi-component intervention categories were identified as impactful tool to reduce food waste in short- but as well in long-term. More randomized controlled trials among various settings with direct food waste measures are necessary in order to promote legislative-evidence based polices. Finally, a harmonization of the methods between and within the different intervention settings would make quantified food waste outcomes better comparable and reliable.

MICROORC: orchestrating food system microbiomes to minimize food waste

Simona Mincione¹, Dr. Patrizia Circelli¹, Manuela Guiducci¹

¹Ciaotech Srl - Gruppo PNO

Aim: Food loss and waste (FLW) pose significant challenges globally, affecting the environment, food security and economies. Approximately, 1.3 billion tonnes of food are lost or wasted annually, which could feed about 1.26 billion hungry people worldwide. In the EU, 14% of food produced for human consumption is lost or wasted, with households and distribution accounting for 70% of this waste. Consequently, reducing FLW has become a critical priority on the sustainability agenda at all levels.

This study aims to showcase the efficacy of microbiome-based tools and solutions in enhancing the microbial quality and safety of protein-rich perishable food products, thereby reducing and preventing food spoilage and waste.

Method: The MICROORC project is designed to develop sustainable solutions for the microbiome monitoring and orchestration by a) developing predictive analytics models incorporating microbiome information to predict shelf-life; b) implementing time-temperature indicators (TTIs), sensors and smart label solutions for dynamic shelf life labelling; c) creating rapid detection assays for microbial indicators of food spoilage; d) enhancing microbiome-based protection technologies to replace synthetic chemicals and increase shelf-life and safety; e) introducing novel packaging solutions targeting spoilage for sustainable development and increased shelf-life.

Three selected products serve as case studies: fresh chicken, smoked salmon, and plant-based meat analogues. These products represent high quality, healthy protein sources with relatively low greenhouse gas emissions and land use.

Results: The results of the study revolve around the optimal combination of sustainable microbiome-based solutions to reduce food waste caused by spoilage, pathogen growth or expiration by 50%, and 10% of products implementing new packaging concepts. By combining microbiome-based packaging technologies with bioprotective strategies that naturally extend the products shelf-life, MICROORC is expected to significantly reduce FLW, potentially saving more than 400 ktons of protein rich food in Europe annually.

Conclusion: The MICROORC methodology not only aligns with the EU and FAO's emphasis on the untapped potential of microbiome-based innovation for transformative change in the food systems, but also offers a strategic and business pathway to improve sustainability and reduce FLW through advanced biotechnology food applications.

Storage characteristics and factors influencing thermo-oxidative stability of cold-pressed blackcurrant, strawberry, and raspberry seed oils

Mgr Yolanda Rajagukguk¹, dr. Mahbuba Islam¹, dr. Anna Grygier¹, dr hab. inż. Aleksander Siger¹, dr hab. Jolanta Tomaszewska-Gras¹

¹Poznan University Of Life Sciences

Corresponding to European Green Deal initiatives, the implementation of the circular economy in the food industry can be fostered by turning waste into resources. Berry seed oil (BSO) is one of the valorised goods from berry by-products. The demand for BSO as a food supplement, nutraceutical, and cosmetic product has been observed in recent years. Despite the growing interest, the consumer remains unprotected from possible fraud because there is no enforceable regulation to control the quality and the authenticity of BSO.

Aim:

This study aims to determine the storage characteristics of cold-pressed blackcurrant, strawberry, and raspberry seed oils and the factors influencing thermo-oxidative stability during storage.

Method:

Each oils was packaged in a brown-glass bottle without headspace, stored at $\pm 20^{\circ}\text{C}$ to mime the store condition for one year. The sampling was conducted at 0, 3, 6, 9, 12 months. Physicochemical, compositional, and thermal stability analyses were carried out every observation months. Factors influencing thermal stability were determined based on step-wise regression.

Results:

After one year, minor degradation was observed as shown in hydrolytic rancidity (acid value), oxidation products (p-anisidine, K232, K238 values), and antioxidant activity (DPPH radical scavenging, tocopherol). Pigment degradation became noticeable after 6 months as observed in chlorophyll, carotenoid, and colour (hue, chroma, ΔE) parameters. No major oxidative deterioration took place in all oils as observed in thermo-oxidative stability (OIT 120, 140 °C) and fatty acid composition, as the remaining tocopherol content is still abundant after one year.

Conclusion:

As a valorised by-product, the initial quality state of BSO heavily depends on the raw material quality prior to oil extraction. Despite the quality changes, berry seed oils still possessed excellent thermal stability after one year. The overall regression results show the negative (chlorophyll and K268) and positive (tocopherol) influences on thermo-oxidative stability of berry oils during storage. The presented works provide a new insight for BSO producers to design a suitable process based on individual characteristics of blackcurrant, strawberry, and raspberry oils during storage.

Non-destructive internal disorder detection of 'Envy' apple by bio-impedance and electric equivalent model.

Ms. Sundus Riaz^{1,2,3}, Mr. Pietro Ibba¹, Miss Nadja Sadar², Mr. Ahmed Rasheed¹, Mr. Stephan Sturz², Mr. Angelo Zanella², Miss Luisa Petti¹, Mr. Paolo Lugli¹

¹Faculty of Engineering, Free University of Bozen-Bolzano, ²Department of Storage and Post-harvest Biology, Laimburg Research Centre, ³Faculty of Agricultural, Environmental and Food Sciences, Free University of Bozen-Bolzano

Aim:

Apple (*Malus domestica*) is one of the most consumed fruit and valuable cash crop globally, because of its great economic importance in European market, that based on customer demand. In this region diverse range of apple varieties are produces, among them some are susceptible to internal browning during cold storage. This study aims to develop an efficient non-destructive technique to estimate internal browning in apples after post-harvest cold storage using electrical impedance spectroscopy (EIS) technique and validated by well-investigated optical NIR techniques.

Method:

For the sake of large variety of data, Envy apple were harvested from two different orchards at two different harvest window. One data sample harvest early and other at late harvest time. Then store all apple in two different storage conditions for seven month. After remove from cold storage all fruits were exposed for 7 days of shelf life (SL) at 20°C and 85% relative humidity (RH). Anlayed fruit sample from equatorial region at both the sunny and shady side of the apple and acquire information about all then changes within the fruit. All sample were analysed by EIS and NIR.

Results:

At low- and mid-frequency region of the bioimpedance spectrum, a significant differentiation was observed between healthy and internally browned samples (p -values ≤ 0.01). The frequency points (40 Hz, 600 Hz, nd 10 kHz) efficiently investigate the physiochemical changes that are produced during the development of internal browning in apple, but these significant pronounced results are not obtained by 1MHz. On the contrary, the measurement of dry matter content did not show promising discrimination between healthy and internally browned apple (p -values ≥ 0.05). Further bio-impedance results were fitted by best fit equivalent circuit model, the acquired circuit parameters (RI, RE, CPE) further validate the results of bio-impedance.

Conclusion:

In future, this non-destructive, portable, cheap, user, and environment friendly technique will help to reduce the post-harvest paradox loss and address sustainable agriculture and food security.

FOLOU: Bringing knowledge and consensus to prevent and reduce Food Loss at the primary production stage

Driss En-Nejjary

FOLOU is a Horizon Europe project focusing on food loss at the primary production stage from multiple perspectives. It introduces advanced methods to measure, monitor, and reduce food losses across Europe, which is fully aligned with the EU Green Deal and Farm to Fork strategies aiming to create a fair, healthy, and environmentally friendly food system.

The project addresses many aspects, challenges, and problems related to food loss at the primary production stage. It will establish a standardized definition of food loss, create food loss registries, provide training on food loss estimation and prevention. Food commodities include grains, fruits, vegetables, and fisheries. FOLOU also promotes innovative technologies for measuring and reducing food loss. Cutting-edge technologies like UAVs, satellites, AI algorithms, Blockchain, Big data, and sentiment analysis to create novel and unified methods are applied and tested to quantify and reduce food losses.

Through a comprehensive and integrated approach that combines research, training, and practical implementation, FOLOU aims to transform food production into a more sustainable and efficient system, reducing environmental impact and strengthening food security across Europe.

Food loss, primary production, sustainability, UAVs, satellites, AI, Blockchain, Big data, Sentiment analysis.

New packaging protocol for hydroxytyrosol-rich black olives with lower acrylamide content.

Mercedes Brenes Álvarez¹, Concepción Romero Barranco¹, Manuel Brenes Balbuena¹, Eduardo Medina Pradas¹, Eva María Ramírez Castro¹, Pedro García García¹

¹Spanish National Research Council (CSIC)

Aim: Olive branches are a by-product that is generated in large quantities in groves and factories. This residue is currently used for multiple purposes, one of them is the extraction of bioactive substances. The aims of this study were (i) to get a polyphenol extract rich in hydroxytyrosol from KOH treated olive branches; and (ii) to assess the effect of this extract on the acrylamide formation, color and phenolic content of packed black olives.

Method: Olive branches of the Manzanilla cultivar were cut into small pieces, covered with 10 g/L KOH and left at ambient temperature for 24 h. Then, the alkaline solution was concentrated to 20% of its initial volume in a rotavapor under vacuum at 60 °C. Seven batches of pitted olives were bottled in A314 jars. The fruits were covered with standard brine or brine spiked with the concentrated alkaline solution to reach 1000 mg/L or 2000 mg/L hydroxytyrosol. Then, the jars were sterilized to reach an accumulated lethality of 15 F₀. Superficial color, phenolic compounds and acrylamide concentration in the final product were analyzed after two months.

Results: The main polyphenol in the concentrated KOH olive extract was hydroxytyrosol (6551 ± 123 mg/L). A low content of oleuropein (1235 ± 28 mg/L) was also detected. The addition of this solution to the canning brine did not have a significant effect on the color of the sterilized product. By contrast, it significantly increased the concentration of phenolic compounds in the pulp of the olives (160 mg/kg of hydroxytyrosol) versus the content in the control sample (29 mg/kg of hydroxytyrosol). The bitter secoiridoid oleuropein was not detected in the canned product. Another benefit could be the reduction of up to 32% of the acrylamide formation during the sterilization step.

Conclusion: Olive leaf extract rich in hydroxytyrosol can be obtained from the KOH treatment of olive branches. The spiked of this extract in the packing brine of black olives did not affect the color of the fruits while it enriched them in hydroxytyrosol. In addition, a reduction in the acrylamide concentration of up to 32% was achieved.

Evaluating Swiss Food Side Streams for Sustainable Food Product Development

Ivana Salvatore¹, Ramona Leue-Rüegg¹, Dr. Claudio Beretta¹, Prof. Dr. Nadina Müller¹

¹ZHAW Zürcher Hochschule für Angewandte Wissenschaften

Aim:

Recognizing the substantial environmental, economic, and moral implications of food waste, this research project focuses on utilizing the potential of food manufacturing side streams, which are generated in substantial amounts and consist of diverse valuable components suitable for valorization. Using side streams in the development of new food products requires a solid know-how on their detailed composition including both valuable components as well as critical contaminants plus technofunctional characteristics. The final aim of this project is to evaluate processing strategies for 12 impactful Swiss food side streams and their application potential of in food products complemented by the assessment of the ecologic and economic effectiveness of the solutions.

Method:

12 side streams were selected based on their environmental impact and the amount generated in Switzerland. Detailed analyses were then conducted to assess the technofunctional, biological, chemical, and nutritional qualities of these streams. Technofunctional analysis includes foamability, emulsifying properties, water absorption amongst others. Furthermore, the feasibility of various upcycling processes was explored, including mechanical, thermal, biological, and chemical treatments, to enhance the side streams' suitability for further applications.

Results:

The investigation identified several side streams with promising characteristics for valorization. The selected agri-food side streams exhibited diverse biological, chemical, nutritional and techno-functional properties, highlighting the complexity and uniqueness of each material. Through targeted processing treatments, the influence on those properties could be observed and valuable insights for further applications could be gained.

Conclusion:

By taking advantage of the valorization potential of underutilized side streams, the project contributes to the advancement of sustainable food production practices.

Process-Structure-Properties relationships during alkaline hydrogen peroxide processing of brewer's spent grains (BSG)

Marcio Sanches², Vitor Stochi², André Borges-Machado², **Pedro Augusto**¹, Tiago Polachini², Javier Telis-Romero²

¹Université Paris-Saclay, CentraleSupélec, Laboratoire de Génie des Procédés et Matériaux, Centre Européen de Biotechnologie et de Bioéconomie (CEBB), ²Food Engineering and Technology Department, São Paulo State University, Institute of Biosciences, Humanities and Exact Sciences (Ibilce)

Aim:

The valorization of agro-industrial by-products is a necessary approach in the context of sustainability, bioeconomy and circular economy. However, a notable gap in the literature concerns the engineering properties of by-products, intermediate and final products, limiting their process design. This work evaluated the changes in composition, structure and properties during alkaline hydrogen peroxide (AHP) processing of brewer's spent grains (BSG) – which constitutes 85% of brewing industry by-products, with a global production estimated in almost 40 million tons yearly.

Method:

BSG from a local brewery were studied, therefore constituting a real industrial by-product. Suspensions with different concentrations of BSG (2–8%) were subjected to different processing times (0–12 h) using AHP (1–8%) at 20°C. The BSG chemical composition, morphology, crystallinity, modifications of functional groups, and rheological behavior were evaluated over processing, as well as the techno-functional properties of the obtained protein-rich fractions.

Results:

AHP process removed proteins, lignin, and extractives from BSG into the suspension, increasing the cellulose and hemicellulose content in the remained structure – which presented a cell wall thinning, changes in particle shape and higher crystallinity. AHP probably reacted selectively with lignin, providing delignification of the biomass, and facilitating the penetration of reagents into the matrix, also improving protein extraction. Those effects increased with both AHP concentration and processing time, being reduced by increasing BSG content. The suspensions presented a Herschel-Bulkley flow behaviour, whose yield stress and consistency index increased over processing due to the compounds extraction to the suspension. The protein extraction yield increased from ~30% to ~50% by applying this process, increasing the foaming capacity and stability, but reducing the water holding capacity of this fraction, with no impact on the oil holding capacity.

Conclusion:

The increased concentration of cellulose and hemicellulose in the processed solid matrix suggests a better substrate for efficient bioethanol/biofuel production, as well as a less reactive and less hydrophilic matrix for bio-based materials production. Moreover, the obtained proteins can be studied as potential source for feed, food or even materials production. The developed processes will be useful for the valorisation of this agro-industrial by-product towards the circular economy.

Incorporation of bioactive compounds from orange peels waste to produce a functional cheese in brine

Dr Marianna Giannoglou¹, Dr Varvara Andreou¹, Ms Zacharoula Maria Xanthou¹, Prof Vasilis Valdramidis², **Dr George Katsaros**¹

¹Institute of Technology of Agricultural Products ELGO-DEMETER, ²Department of Chemistry, National and Kapodistrian University of Athens

Aim: Efficient fruit waste management is crucial for environmental sustainability, economic well-being, and regulatory compliance. Orange peel waste is rich in bioactive compounds, including flavonoids, carotenoids, and phenolic acids, which are well-known for their antioxidant properties and potential health benefits. The extraction and incorporation of these compounds into a variety of foods may improve their functionality. The research aimed to utilize bioactive compounds extracted from orange peel waste in the production of an innovative white cheese in brine, enhancing its functional properties while ensuring optimal quality.

Method: A powder of bioactive compounds (total phenolics: 29.25 mg/g dm, antioxidant activity: 32.90 mg/g dm) was received after ultrasound processing of orange peel wastes using a green solvent solution of ethanol/water 50/50 v/v. Four different amounts of the bioactive compounds powder (0.5-2%) were incorporated into ovine milk (5.8% fat content, 12.2% total solids) and four cheeses were manufactured according to Feta cheese-making and ripening conditions. One month after ripening at 4°C, microbial, quality, and sensory analyses of the products were performed.

Results: The increase in the concentration of powder in milk resulted in increased concentration of bioactive compounds in cheese. However, a significant amount of these compounds was removed along with the serum during syneresis process. The increase in the total solids of the milk resulted in cheeses of significantly increased consistency. Based on a comparative evaluation of the received data derived from the sensory evaluation, texture analysis and cheese antioxidant analysis, an incorporation of 1.5% of the powder in milk was considered as the optimum amount. The activity and viability of the starter culture was not affected by the addition of powder in milk. The final produced cheese was of superior quality and of increased antioxidant activity by 17% compared to Control, while this cheese received higher organoleptic scores compared to the control sample.

Conclusion: Fruit waste, such as orange peels, has a lot of potentials to be used as sources of functional ingredients for high-quality final food products. This sustainable approach not only reduces waste but also promotes resource efficiency and culinary innovation.

Upcycling a local by-product: characterisation of “Verjuice” and application as acidifier in elderflower syrup.

Msc Annika Kofler¹, Flavia Bianchi¹, Giulia Maria Marchetti¹, Niccolo Trentini¹, Elisa-Maria Vanzo¹, Elena Venir¹

¹Laimburg Research Centre

Aim: Verjuice, extracted juice from unripe grapes, is a by-product of viticulture obtained by upcycling the grapes harvested during the thinning stage and thereby preventing waste. The high total acid content, very low pH and sour taste of the grapes make this juice a sustainable acidifying ingredient, and a suitable substitute for acidifying additives. The aim of this study was to assess the potential benefits of verjuice as a natural acidifier in elderflower syrup. Traditional elderflower beverages are popular due to their pleasant taste and pro-health properties. Elderflower syrup has been reported to be a good source of phenolics, contributing to the prevention of diseases caused by oxidative stress. Elderflower syrups produced with the verjuice and with a commonly used acidifier (citric acid), were compared and data are discussed with regards to chemical characterization, antioxidant activity and sensory characteristics.

Method: The total polyphenol content, measured with the Folin-Ciocalteu assay; ascorbic acid content, measured with Ascorbic Acid Test Kit; and antioxidant activity, assessed using the DPPH bleaching rate, were analysed in elderflower syrup made with both, verjuice and citric acid. The sensory analysis was carried out using a tetrad test, to identify perceptible difference between the two samples.

Results: Elderflower syrup produced with citric acid showed a slightly higher amount of total polyphenols (390±12 mg/L GAE) compared to the syrup with verjuice (329±8 mg/L GAE). Also, ascorbic acid contents were slightly higher in the samples produced with citric acid (28±4 mg/L with citric acid, 21±3 mg/L syrup with verjuice). Interestingly, a significantly higher antioxidant activity was measured in the Syrup with verjuice resulting in a Dpph bleaching rate of 0.6 ±0.07 OD515min-1µgDM-1 and 0.4 ±0.06 OD515min-1µgDM-1 in the syrup with citric acid.

Conclusion: Acidifying additives can be replaced with verjuice in elderflower processing, leading to a clean label product, and a slight improvement of the nutritional value of the final food, in terms of a significantly higher antioxidant activity. Increasing the range of application of verjuice supports the circular economy and the use of local ingredients.

Factors affecting the phenolic composition of olive leaf infusion

Dr. Eduardo Medina Pradas¹, Dr. Eva M. Ramírez Castro¹, Mercedes Brenes Álvarez¹, Dr. Manuel Brenes Balbuena¹, Dr. Concepción Romero Barranco¹

¹Instituto de la Grasa - CSIC

Aim: Olive leaf, a byproduct of the olive industry rich in beneficial phenolic compounds such as oleuropein, remains not fully exploited in healthy infusions. This study aims to (i) investigate the evolution of phenolic compounds during storage of dried leaves, (ii) assess the influence of infusion time and temperature on polyphenol extraction, and (iii) evaluate acceptability through a consumer panel test.

Method: Olive leaves from Hojiblanca and Manzanilla cultivars were harvested and stored for 24 hours before being dried at 80°C using infrared drying equipment for 35 minutes. The polyphenol content of dried leaves was characterized by HPLC over one year, with samples stored at room temperature both whole and ground. Infusions were prepared using water at temperatures ranging from 4°C to 100°C for 1 to 20 minutes of maceration periods. A sensory evaluation of bitterness and overall acceptability was conducted by a panel test.

Results: The concentration of polyphenols in dried olive leaves ranged from 10 to 12 g/kg, remaining stable throughout one year of storage regardless of leaf form. Optimal conditions for infusion preparation, maximizing phenolic compound extraction, were found to be 100°C for 15 minutes.

Regarding the sensory panel evaluation, bitterness was noted in infusions containing high oleuropein levels, reaching an acceptability threshold of 144 mg/L. However, the acceptability increased when infusions were sugared, tolerating higher oleuropein concentrations up to 212 mg/L.

Conclusion: This study successfully optimized the infusion of olive leaves to retain the highest concentration of oleuropein, known for its health benefits, providing consumers with infusions rich in this compound but non-bitter.

Figuras posibles para el poster.

Figure 1. Curvas de secado

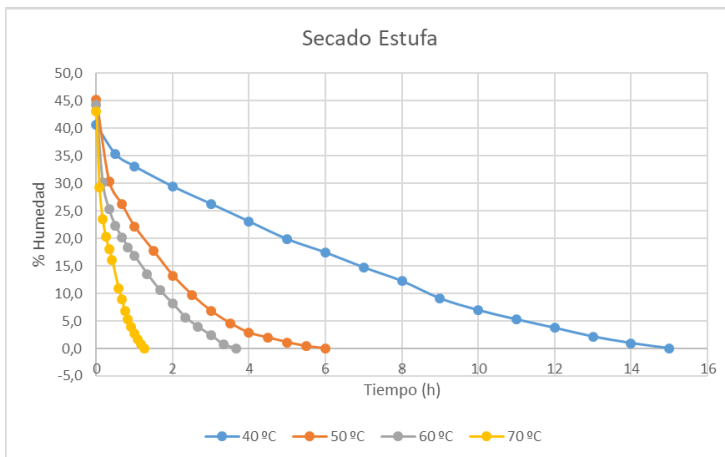
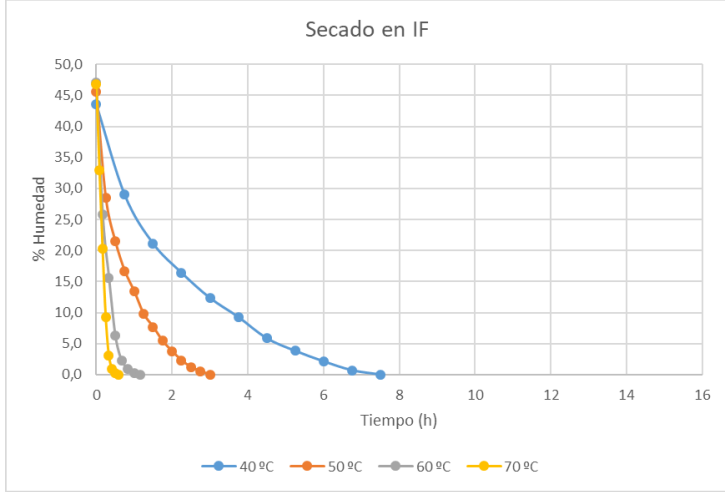


Figura 2. Ecuación de Arrhenius

Modelo de page: $MR = \exp(-ktn) + b$ para $n=2$

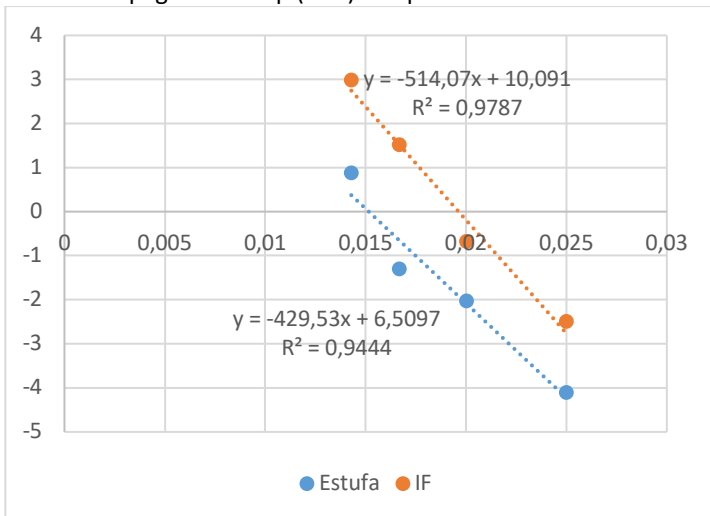


Figura 3. Concentración de fenoles en la hoja antes y después del tratamiento térmico expresado en peso seco (ppm).

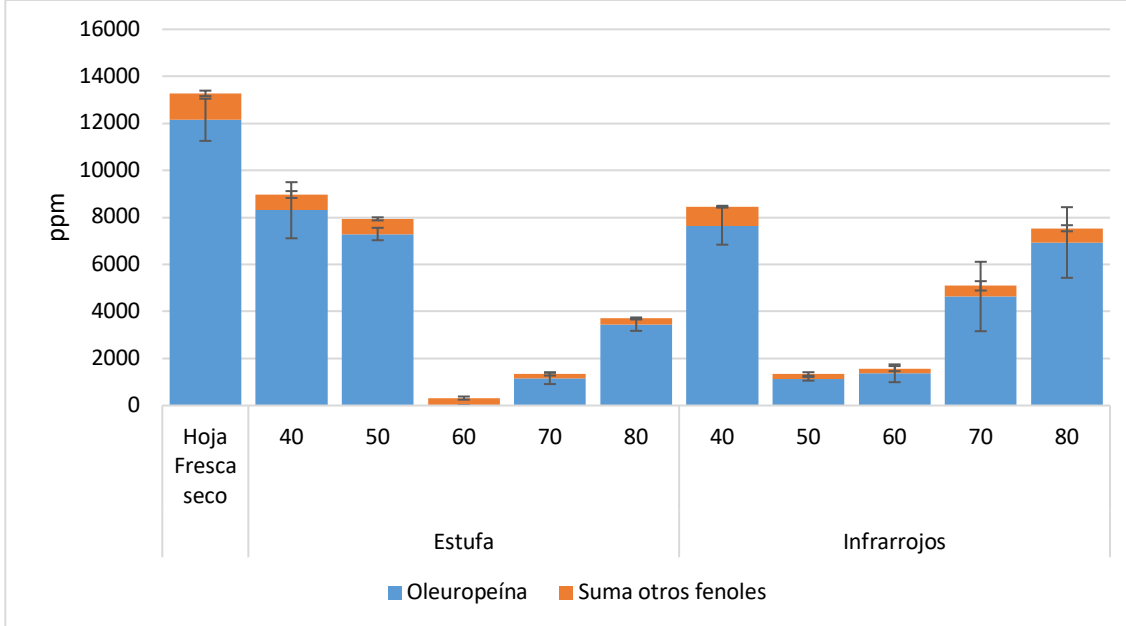


Figura 3. Concentración de fenoles totales en la hoja antes y después del tratamiento térmico expresado en peso seco (ppm). Secadas el mismo día o mantenidas a temperatura ambiente 24 horas.

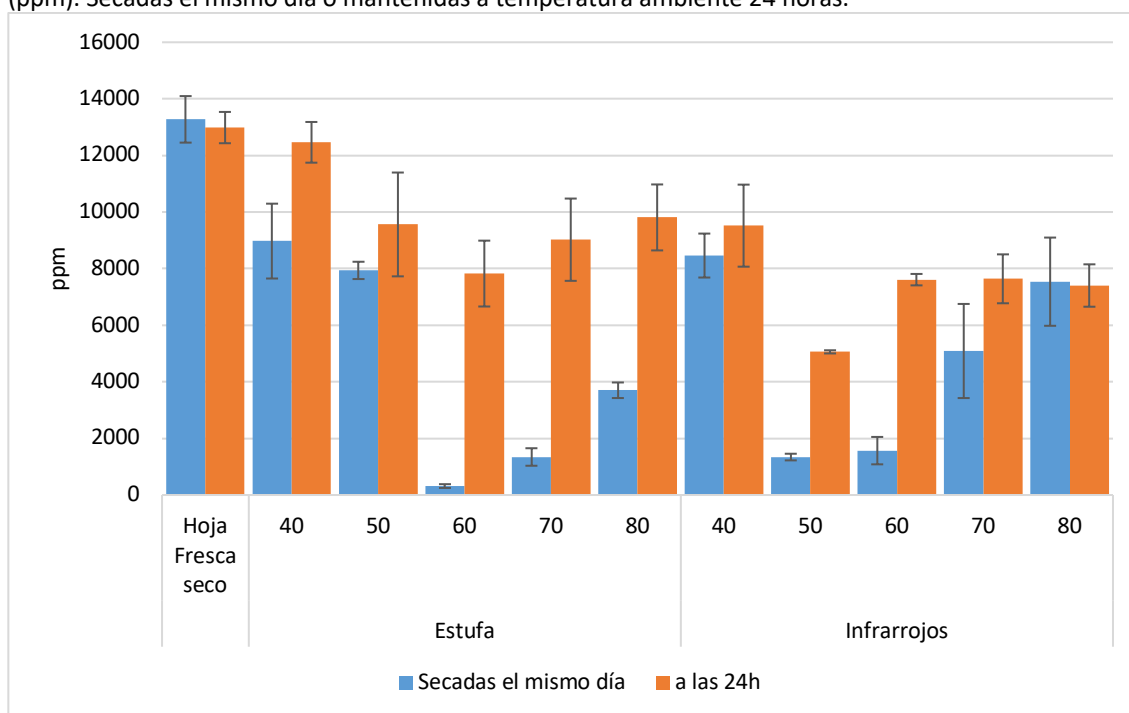


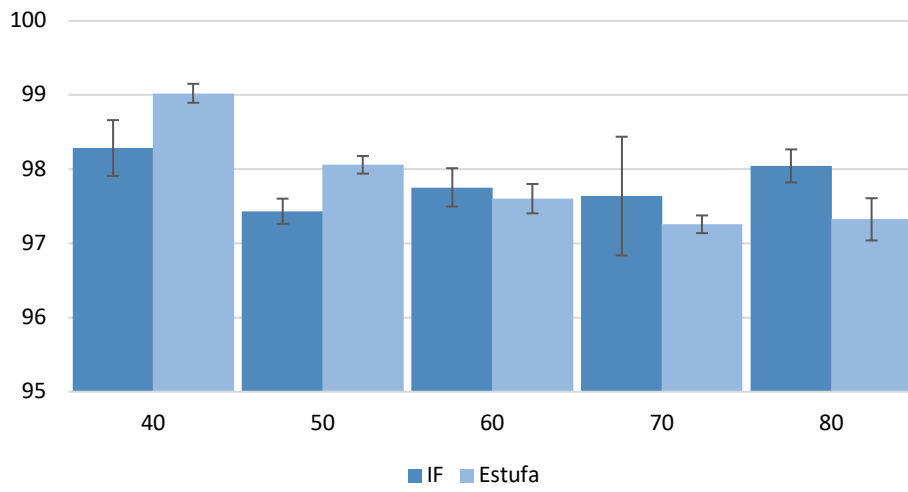
Tabla 1. Concentración de Oleuropeína en la hoja y en la infusión.

	Oleuropeína ppm en la hoja		Oleuropeína (ppm) Infusión		
	Media Ole	SD Ole	Media	SD	
Estufa	40	8306	1194	377	27
	50	7293	264	271	3
	60	0	0	0	0
	70	1152	240	49	5
	80	3428	252	192	9
Infrarrojos	40	7632	791	378	23
	50	1136	79	67	2
	60	1370	378	71	0
	70	4637	1476	186	1
	80	6934	1502	358	7

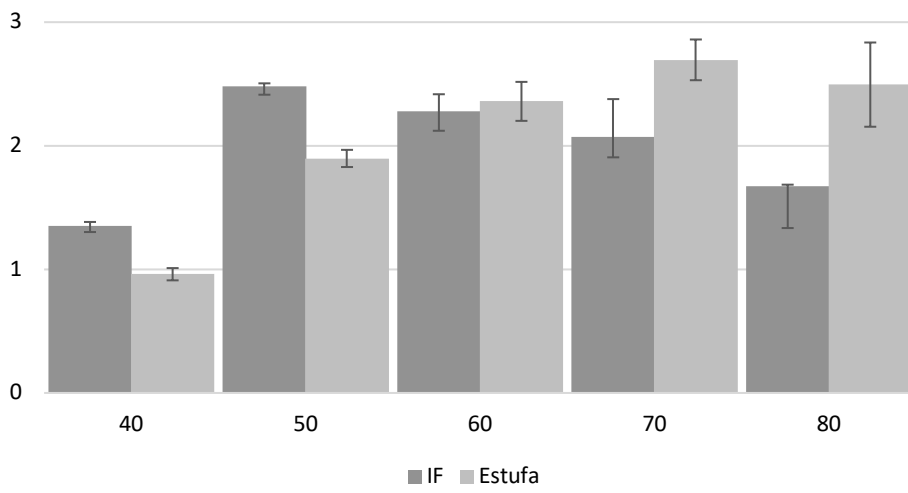
Fotos de equipos de IF y estufa.

Figura. Color de las infusiones

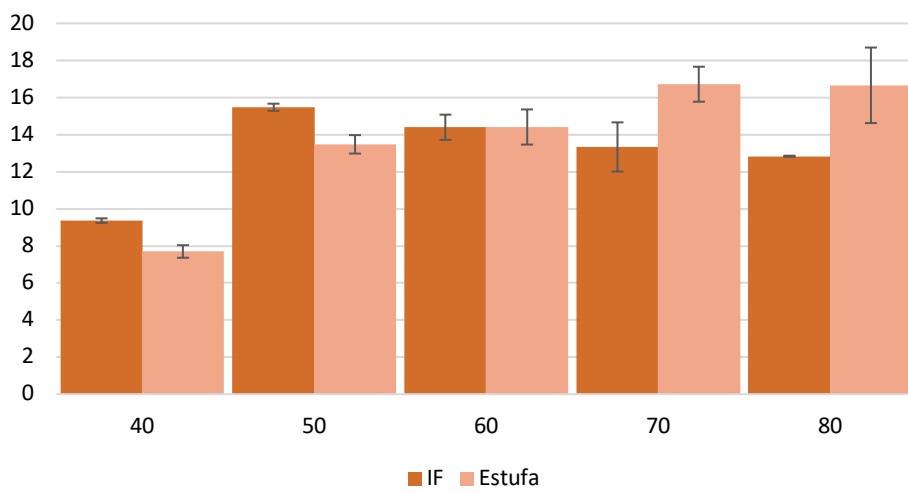
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a*



a*



Fresh goat cheese enriched with date paste: a novel ingredient for the dairy industry

Clara Muñoz Bas¹, Nuria Muñoz Tébar¹, Raquel Lucas González¹, Manuel Viuda Martos¹, Juana Fernández López¹, Estrella Sayas Barberá¹, José Ángel Pérez Álvarez¹

¹Miguel Hernandez University

Aim: The aim of this work was to analyse the effect of date paste addition on physico-chemical and microbiological properties of fresh goat cheese.

Method: Date paste (DP) was obtained from coproducts of date fruits (Confitera cv.) at the tamar ripening stage. Fresh goat cheese was prepared with pasteurized goat milk (72 °C for 15 s). Three vats (20 L milk each one) of cheese including the control vat and 2 vats enriched with DP (4 and 8%) were processed. After the addition of 0.05 Danisco culture units (DCU)/L of the starter culture (CHOOZIT MA4001; Danisco, Sassenage, France), date paste was added to milk and stirred for 10 min followed by the addition of 1 mL/ L of rennet (calf rennet, Laboratorios Arroyo, Cantabria, Spain,). Fifteen pieces of cheese (~0.25 kg) were obtained from each batch and stored in a chamber until use at 5 °C and 85% relative humidity. Physicochemical parameters (pH, Aw, color CIELAB* and texture (TPA) and microbiological quality (*Lactobacillus spp* & *Streptococcus spp*, enterobacteria, molds and yeast, and total aerobic bacteria) were assessed in triplicate.

Results: The addition of DP didn't impair either the cheese-making process or coagulation. Regarding the physicochemical properties, significant differences were observed in the pH (control: 5.73 ± 0.04 ; 4%: 5.43 ± 0.02 ; 8%: 5.57 ± 0.03) but the aw remained similar ($p > 0.05$) among the three batches. However, significant differences were observed in the coordinates L*, a* and hue (h). With regards to the textural parameters of the cheeses, significant differences ($p < 0.05$) were observed in hardness, adhesiveness, springiness, gumminess and chewiness, while no significant differences were noticed in cohesiveness and resilience. For the microbiological quality, enterobacteria were not detected in any sample while very low counts were observed for molds and yeast in the cheeses containing the DP. Total aerobic bacteria decreased whilst lactic acid bacteria significantly ($p < 0.05$) increased when DP was incorporated.

Conclusion: The use of date coproducts for the enrichment of fresh cheese is technologically feasible without adversely affecting the coagulation process. The addition of date paste in fresh goat cheese enables to obtain products with physicochemical properties and viable microorganisms counts similar to those of plain fresh cheese.

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Innovative formulation of added value spreads from oil cakes

Lidija Perović¹, Branislava Đermanović¹, Miloš Županjac¹, Milica Stožinić², Ivana Lončarević², Aleksandar Marić¹, Jelena Tomić¹

¹Institute of Food Technology in Novi Sad, ²Faculty of Technology Novi Sad

Aim: Oil cakes represent a by-product from the oil industry left after cold-pressing oil from the seeds. Compared to the most popular conventional raw materials, oil cakes, as a source of various nutritionally valuable compounds, offer a competitive edge for use in the development of plant-based spreads. This is particularly pertinent in light of the growing consumer preference for plant-based foods, whereby the utilization of these by-products can improve the nutritional value and health benefits of well-known foods. Following the concept of zero waste and circular economy, the main goal of this research is to create nutritionally valuable spreads by using various oil cakes.

Method: Nutritionally valuable spreads are going to be produced in ball mill. Conventional technique will be used for operation with unconventional raw materials used in a completely new manner. The appropriate formulations will be designed to embrace an optimum balance between the pursued target, manufacturing issues and sensorial characteristics of the final products. Spreads are going to be evaluated under sensory tests, texture analysis, particle size distribution and oxidative stability.

Results: Innovative spreads made of protein rich oil cakes, left after pressing oil from seeds are rich in fibre and protein and have acceptable sensory profile and particle size distribution. Moreover, the use of unconventional raw materials will aim to produce spreads that can carry the nutritional claims of "high fiber" and/or "source of plant protein. After cold pressing oil from the hazelnut and walnut seed around 44-45% of oil-cake (by-product) is left. Obtained by-product is raw material for spreads formulation. In the walnut cake after pressing oil still remains 15% of the oil and also contains 45% of proteins and 20% fibres. Nutritionally valuable spreads are enriching offer of healthy food.

Conclusion: Currently, society is searching for new plant-based sources of proteins, due to health consciousness and observing for sustainable diet. By-product left after production of the main product is an ecological problem that must be solved. By creating spreads with by-products are reaching global world goal being in circular economy and unlocking new way of reaching zero waste concept.

Whey-based fermented milk products as an alternative to whey processing in artisanal cheese dairies

M.sc. Josephina Scherbanjow¹, Dr. Urte Schleyerbach¹, Prof. Dr. Sabine Bornkessel¹

¹University Of Applied Science Osnabrück

Aim: For artisanal dairies, processing of sweet whey often poses distinct challenges. Unlike industrial milk processing, the energy-intensive processing of whey, e.g. into whey powder, does not appear to be worthwhile in artisanal production due to small quantities of produced sweet whey and the technical effort required for such processing. However, from a nutritional perspective, sweet whey contains ingredients that can contribute to a balanced diet and therefore its use can have a positive impact on health.

The aim is to develop a sweet whey product that can contribute to human nutrition and can be easily implemented in the daily work of artisan cheese dairies from a technological and work management point of view.

Method: Based on an online survey of 66 artisan cheese dairies in Germany on their use of sweet whey, a recipe for three whey-based fermented milk products (yoghurt, soured milk and kefir) have been developed. The recipes will then be the subject of a field test in a selection of artisan dairies to determine whether the recipes can be implemented in practice. As an example of the other fermented milk products, a nutritional analysis of yoghurt was carried out.

Result: Trials to develop the whey yoghurt, soured milk and kefir recipes have shown that up to 30% of the used milk in traditional recipes can be replaced with sweet whey without high differences in texture and taste compared to traditional products. These products are easy to implement in artisanal dairies that already produce fermented milk products and may have a positive impact on health due to the inherent ingredient composition.

Conclusion: Due to its production method, whey-based fermented milk products can be an economically and ecologically sustainable solution for the use of sweet whey in artisanal cheese dairies. However, further research is needed to determine the consumer acceptance.

Effect of Storage Conditions on Brewer's Spent Grain: Implications on Quality, Safety and Food Application

Pramod Pandurang Aradwad^{1,4}, Muhammad Tayyab¹, Dr. Sharvari Raut¹, Dr. Ahmed Abdelfattah¹, Prof. Dr.-Ing. habil. Cornelia Rauh², Prof. Dr. agr. habil. Barbara Sturm^{1,3}

¹Leibniz Institute for Agricultural Engineering and Bioeconomy, ²Technische Universität Berlin, Straße des 17, ³Albrecht Daniel Thaer Institute for Agricultural and Horticultural Sciences, Humboldt-Universität zu Berlin, ⁴Indian Council of Agricultural Research-Indian Agricultural Research Institute

Aim: The beer industry produces 38 million tons/year of brewer's spent grain (BSG) worldwide. Currently, BSG is being considered, not only as animal feed, but also as alternative food for humans. BSG is not immediately used, as it undergoes a transitional phase after post-production at breweries. Therefore, effective storage is a crucial step not only for preserving quality and safety but also for providing adequate time for the further processing of highly perishable and microbial prone BSG material.

Method: Based on preliminary test, an analysis period of 12-hour was set. In this study, we investigated the influence of storage conditions at 4°C, 20°C, and 35°C for 240, 60, and 36 h, respectively, on the physiochemical properties and microbiological diversity of BSG. Both fresh and stored BSG samples were subjected to quantitative and qualitative analysis using standard analytical techniques.

Results: The results showed that storage temperature and duration significantly influenced BSG properties, with higher temperatures accelerating color degradation (L^* : 50-36, a^* :5-8 and b^* :25-20), moisture loss, and the development of off-odors, stickiness, and fungal growth. Samples stored at 4°C showed favorable attributes, while samples stored at 20°C and 35°C showed significant changes indicating spoilage with respect to storage duration. Chemical analyses indicated minor to moderate changes in protein, fat, starch, fiber, total sugar and phenol concentrations with respect storage temperature and duration. GC-MS identified 25-30 VOCs, mainly alcohols, acids and esters with different storage temperatures yielding distinct compounds. Fresh and samples stored at 4°C exhibited sweet, fruity, and fresh notes, while those at higher temperatures exhibited sour, pungent, and rancid odors, with higher likely originating from enzymatic oxidation and maillard reactions. Changes in storage temperature and duration can profoundly influence the transformation of compounds within BSG, consequently influencing its aroma profile and resulting flavor. Microbial growth rates correlate positively with temperatures (20, and 35°C) and increasing storage periods.

Conclusion: These findings underscore the importance of storage conditions in preserving BSG quality. A comprehensive understanding of the dynamic changes in BSG's chemical, microbial, and flavor profiles during storage is crucial for ensuring its suitability for development of various food products.

Tracing software ecosystem for food and feed utilising the Universal Traceability data eXchange format

Dr. Alexander Falenski¹, Marco Rügen¹, Dr. Olaf Mosbach-Schulz², Professor Dr. Annemarie Käsbohrer¹, PD Dr. Bernd-Alois Tenhagen¹, Dr. Marion Gottschald¹

¹German Federal Institute For Risk Assessment, ²European Food Safety Authority

Aim:

In response to the increasing globalization and complexity of agri-food chains, robust interoperable tracing software tools and streamlined tracing data exchange within the software ecosystem from farm to fork are necessary. A collaborative research initiative between the European Food Safety Authority (EFSA) and the German Federal Institute for Risk Assessment (BfR) is currently underway. This project aims to develop a workflow for tracing data exchange and introduces the Universal Traceability data eXchange (UTX) format. This initiative aims to enable the exchange of tracing data between the various existing software tools for tracing data collection, analysis and reporting via the UTX format.

Method:

Essential as well as accessory variables for tracing back foodborne disease outbreaks along the supply chain were collected and grouped into categories belonging to three major topics: Stations (food business operator, station (companies, manufacturers, supermarkets), activity), product (product, lot, logistic unit, traceable resource unit) as well as information (investigation, information source, contacts, registration schemes). These variables were collected in a spreadsheet, which was then used as basis for the development of a UTX portal demonstrator using RShiny.

Results:

The UTX format harmonizes essential tracing data elements while simultaneously accommodating data specific for certain software tools, thereby ensuring the preservation of data which might be needed in a later step of the tracing investigation. In addition, a UTX portal is being developed, facilitating the structured collection and extraction of unstructured tracing data, for example from notifications within the Rapid Alert System for Food and Feed (RASFF) as a UTX file. UTX data files might be shared among actors in the food safety system via the RASFF system, streamlining manual data extraction during crises.

Conclusion:

This advancement aims to improve the quality, speed, and comprehensiveness of tracing data, facilitating quick analysis and solving complex foodborne crises more efficiently.

Compensation strategies for the development of food composition predictive models robust to external variations

Phd Elena Fulladosa¹, Magdalene Chong², Andrew Parrott², Alison Nordon²

¹IRTA, ²University of Strathclyde

Aim:

Low-cost devices intended for consumers based on near infrared (NIR) spectrometry may help adoption of sustainable healthy diets. However, these devices should be used at supermarket level and require predictive algorithms robust to external variations. The aim of this work was to evaluate different data analysis strategies to develop robust predictive models for food composition when using spectrometric data subjected to external variations, specifically temperature and packaging material, acquired using low-cost sensors.

Method:

Two low-cost handheld NIR-based devices with different spectral ranges and resolutions were used: the smartphone-based SCiO spectrometer (Consumer Physics Inc., Israel) (device 1) and a handheld low-cost NIR setup developed in house (NIRTA 3.0) that uses a Hamamatsu sensor (Hamamatsu Photonics C14384MA-01, Japan) (device 2). The food matrix samples used in this study were obtained from different anatomical muscles of commercial dry-cured ham selecting regions with minimum visual variation and a uniform appearance. Two types of films commonly used for packing in the food industry were chosen to study spectral variations produced by packaging. Spectra were acquired at different temperatures with and without different packaging materials using device 1 and/or 2. The usefulness of global modelling (GM), generalised least squares weighting (GLSW), loading space standardisation (LSS), and multiplicative effects model (MEM) were explored. The effect of the normal sample's heterogeneity (caused by thin fat streaks or colour variations usually present in dry-cured ham samples) was also evaluated.

Results:

Results show that the inherent food sample heterogeneity produces as much spectral variability as temperature and packaging materials. For temperature compensation, LSS did not decrease the predictive error caused by this factor probably due to the heterogeneity of the samples used. In contrast, the GLSW method decreased the salt predictive errors from 0.52% to 0.46% for salt and from 2.10% to 1.40% for water when using device 1, obtaining similar errors to those obtained when using only spectra acquired at 15 °C. Only a slight effect of packaging was observed, and GM models were found to be the best strategy to compensate it, showing a decrease of bias from -1.35 to 0.012.

Conclusion:

The examined compensation strategies could facilitate the deployment of low-cost spectrometers for consumer use, as they offer an effective means to mitigate or eliminate variations from any source in the data that are unrelated to the properties of interest.

ALTPROTON: An ontology for integration and interoperability of multi-aspect data of protein food production chains

Nilanka Kasthuri Arachchilage¹, Deepak Rastogi¹, Gerard Marrugat², Cristina Fernandez-Avila³, Anita Bhaitha^{1,4}, Lais Speranza⁵, Mercedes Caro², Primrose Chikohomero¹, **Prof. Sergiy Smetana**^{1,6}

¹German Institute of Food Technologies (DIL e. V.), ²AZTI, Food Research, Basque Research and Technology Alliance (BRTA), ³IRIS Technology Solutions S.L., ⁴GreenDelta GmbH, ⁵GreenCoLab – Associação Oceano Verde, ⁶Institute of Food Quality and Food Safety, University of Veterinary Medicine Hannover, Foundation

Aim:

Ontologies provide logical meaning to data and possibility to develop machine-readable data formats. Existing ontologies have captured a significant amount of knowledge in food science domain. However, unattended areas still exist. The present study has aimed to address gaps in existing food ontologies and lay the foundation for a single cloud-based data platform tailored to address complexities of the protein food production chain ensuring the interoperability of multi-aspect data generated within the domain.

Method:

The present study covers conceptual ontology structure development. Ontology specification was completed based on inputs from food industry experts, research centers, and universities involved in the EU-funded project GIANT LEAPS. Identification of key concepts and relations, development of definitions, logical connection of domain knowledge to develop ALTPROTON hierarchical structure, extraction of classes from existing ontologies, thesauruses, and openly accessible databases to introduce ontological interoperability and knowledge reuse, consisted of the development process. The selected relationships referred to hierarchy relations, process flow relations, and attribute possessions among entities within the ontology.

Result:

ALTPROTON ontology contained 326 classes, extracted from existing ontologies and newly developed classes which accurately and consistently describe core concepts of the domain. It connected three main domains of the protein food production chain; quality, sustainability, and food processing via a foundational structure developed to address metadata of protein sources. The most abstract concepts in each domain such as nutritional quality, food safety, allergenicity, sensory and techno-functional properties, and environmental, economic, and social sustainability were addressed using classes and logical relationships to generate a comprehensive, complete profile of a protein food production chain. ALTPROTON ensured interoperability through standardized structure and vocabulary, alignment with external ontologies, and logical domain connection.

Conclusion:

ALTPROTON specifies all key aspects in the protein food production chain, enabling interoperability among diverse systems and integrating multi-aspect data generated in the domain. It provides the foundation for a single data platform, increasing the possibility of logical data storage, mutual operation of the data generated within the protein food production chain, and use of software-driven intelligence to support decision-making while avoiding creation of research data silos and facilitating knowledge reuse.

Unveiling the Link Between Animal Welfare and Broiler Meat Quality Through Muscle Proteomics

Phd Candidate Seren Yigitturk, Ingrid C. de Jong², Shai Barbut^{3,4}, Vincenzo Fogliano¹, Sara W. Erasmus¹

¹Food Quality and Design, Wageningen University & Research, P.O. Box 17, 6700 AA, ²Wageningen Livestock Research, Wageningen University & Research, De Elst 1, 6700 WD, ³Department of Food Science, University of Guelph, Guelph N1G 2W1, ⁴Adaptation Physiology Group, Wageningen University & Research

Aim: Poultry meat is projected to constitute 40% of the protein consumed globally from meat sources. The metabolic functions of animals are influenced by extensive husbandry factors, with consequent impact on animal welfare and intrinsic meat quality. However, the extent to which the extensiveness of animal husbandry affects intrinsic and extrinsic meat quality still needs to be confirmed. This study aims to elucidate texture, color, and water-holding biochemistry in chicken meat and clarify the function of proteins during muscle-to-meat conversion. We hypothesize that animal welfare linked to extensification factors changes the proteome of the *Pectoralis major* (PM), leading to differences in meat quality.

Method: A total of 3680 two different broiler breeds (Hubbard S757N, slow-growing vs. Hubbard JA787, fast-growing) were raised in a higher welfare system enriched with and without lucerne in a net, perch and dust bath with peat in medium-growing space (30 kg/m²). A total of 160 broilers (2 breeds* 2 enrichment, n=5 pens/treatment, n=8 broilers/pen) were randomly selected for animal welfare assessment and meat quality analysis. Meat pH, color, texture and water-holding capacity were measured as related to intrinsic meat quality. Twenty right PM (1 representative broiler/pen) were selected for comparative proteome analysis of which the whole muscle lysate was processed with Protein Aggregation Capture on Microparticles using trypsin. Peptides were measured using nanoLC-MS/MS and quantified by label-free quantification method.

Results: Our findings indicate that slow-growing breed has the most beneficial effects on animal welfare which has a strong impact on the meat quality parameters. The effect of enrichment was only observed in the color parameter (b* value, yellowness) of the intramuscular steak ($p < 0.05$). A total of 1616 proteins were identified, with 216 and 70 proteins differentially expressed ($p < 0.05$) between breeds and enriched conditions, respectively. The implications and biological meaning of these proteomic differences will be discussed.

Conclusion: This is the first study linking extrinsic factors in farm and intrinsic meat quality with muscle proteome in a larger sample size. Results show the pattern of differentially identified proteins can be used to optimize the meat quality ensuring the authenticity of the husbandry factors.

Shaping the future of the food chain: envisioning an Intelligent Food Assurance System

Ms Jiaqi Zhou¹, Prof Paul Brereton¹, Prof Katrina Campbell¹

¹Queen's University Belfast

Aim:

An Intelligent Food Assurance System (IFAS) represents a future "smart" system, set to be integrated with state-of-the-art sensor and digital technologies. The design of IFAS aims to extract valuable information from the vast amount of data within the supply chain by employing various numerical models. This study constructs a food assurance model based on the food risk matrix, incorporating elements such as food quality, safety, authenticity, and defence.

Method:

The design concept of an IFAS is elaborated by first outlining the fundamental concepts and definitions of food systems, and then providing a comprehensive review of recent advancements in technology and intelligent systems.

Results:

The literature review has identified that although numerous studies have applied digital technology to food assurance, there remains gaps in the utilization of data at different locations in the supply chain, particularly in providing a rapid response to food risks.

Conclusion:

In conclusion, the design of IFAS will emphasize the elimination of information gaps between stakeholders by using the common system to help different departments of the supply chain work together more effectively to identify and address risks. Furthermore, the potential influence of an IFAS on enhancing transparency and traceability in the food chain, reducing loss and waste to promote sustainable development, and enhancing customer trust is emphasised. Government may also consider taking on critical tasks such as regulation and oversight to ensure the successful implementation and scalability of IFAS.

Towards a European Foodome: pilot in Hungary

Michael Sebek¹, Milán Jánosov², Péter Ruppert¹, **Erika Országh**³, László Barabási¹

¹Network Science Institute and Department of Physics, Northeastern University, ²Geospatial Data Consulting, ³Syreon Research Institute

Aim:

The main global public health problems are cardiovascular diseases, cancers, obesity and diabetes. These illnesses are responsible for the death of around 100,000 people every year in Hungary alone. A significant part of these leading causes of death can be traced back to environmental and lifestyle factors. The pivotal role of diet has been well supported by nutrition-related research, however, our current understanding of the way biochemicals in food affect health is largely limited to a few hundred components tracked by different food composition databases. These food components represent only a tiny fraction of the total composition of the food supply. This very incomplete knowledge of the biochemical composition of foods hinders research from discovering the mechanistic effects and roles of the thousands of molecules. To solve this problem, we propose a Big Data and AI strategy for the creation of a high-resolution collection of the biochemical composition of foods.

Method:

Data from existing food composition databases (e.g., USDA, FooDB) were connected and curated. The data were complemented by a Machine Learning (FoodMine) assisted literature review and data extraction process focusing on most important Hungarian food commodities. Moreover, compositional data were connected to various health effects data (e.g., CTD) as well.

Results:

A unified database for Hungarian food products (Foodome Knowledge Graph – HKG) have been created. HKG shows the interplay between foods, their compounds, and the potential downstream effects of their consumption. HKG enables connecting food compounds to potential mechanisms of action, both marker and therapeutic, and disease associations as well.

Conclusion:

The complete chemical composition of the most important food products in Hungary is available through HKG, enabling the research and start-up communities to utilise it. This could change the direction of food and nutrition research as well as could set out the future development direction in the health and agricultural industry.

Reproductive toxicity of Bisphenol A (BPA): Multilevel meta-analysis and dose-response analysis

Xin Wang¹, Rajat Nag¹, Nigel Brunton², Abu Siddique², Sabine Harrison², Frank Monahan², Enda Cummins¹

¹School of Biosystems and Food Engineering, University College Dublin, ²School of Agriculture and Food Science, University College Dublin

Aim:

Bisphenol A (BPA) is a widely used synthetic compound frequently detected in many food products. Dietary exposure to BPA is a potential health concern. Adverse effects on the reproductive system from BPA exposure have been reported in animal studies. Therefore, this research aimed to summarise and analyse reproductive toxicity endpoints from BPA exposure in rodent models.

Method:

Articles published in English between 2012 and 2021 were collected from online databases, viz. Scopus, EmBase, Web of Science, and PubMed. Only primary rodent studies investigating BPA exposure from the oral route with multiple dose levels on reproductive toxicity effects were considered. Standardised mean difference was the selected effect size for the multilevel meta-analysis and subsequent dose-response analysis. In addition, the assessment of Risk of Bias (RoB) for individual selected studies and publication bias was conducted. All statistical analyses were performed using open-source RStudio packages.

Results:

The literature search identified 41 studies for the quantitative analysis. Summary effects indicated that oral exposure to BPA significantly decreased sperm concentration (Hedges' g : -1.35) and motility (Hedges' g : -1.12). There were no significant effects on the absolute/relative weight of male (i.e., testis, epididymis) and female (i.e., ovary, uterus) reproductive organs. The toxicological reference values of 0.0011 and 0.0033 mg (kg bw)⁻¹ day⁻¹ were proposed for BPA exposure on sperm concentration and motility based on dose-response analysis.

Conclusion:

In summary, the results highlighted potential health risks associated with BPA exposure concerning reproductive toxicity, supporting the re-evaluation of health-based guidance value (HBGV) by the European Food Safety Authority (EFSA). This study paves the way for a more informed BPA dose response for subsequent use in food risk assessment studies.

Comparison between chemometric analysis and machine learning for the prediction of macronutrients in fresh cheeses

Mercedes Bertotto¹, Esther Kok¹, Meeke Ummels¹, Hajo Rijgersberg¹, Guido Camps¹, Rosalba Calvini²

¹Wageningen University & Research, ²University of Modena and Reggio Emilia

Aim: Automated detection of macronutrients in food could be facilitated by use of machine learning, but currently traditional chemometrics methods are still the baseline method of choice. This study compares traditional chemometric methods with machine learning (ML) methods on how well they predict protein and fat content in a variety of cheeses.

Method: In this study, hyperspectral imaging data (in the 941.1-1723.94nm spectral range) of 76 cheese samples were processed and analyzed to predict their fat and protein content. After calibrating the images and segmenting cheese samples from the background, chemometric and ML analysis were conducted using R and Python, respectively. For both approaches data was split randomly into training and evaluation sets. Next, PLS regression models were trained on the calibration set, tested on the validation set and evaluated in terms of RMSEP and R². Several preprocessing (SNV, derivatives, EMSC) and feature selection methods (CovSel, IPW-PLS, UVE-PLS) were compared. ML results were achieved based on multilayer perceptrons (MLP) on full spectral data, no variable selection. To compare with chemometrics, similar preprocessing steps were performed.

Results: For protein prediction using a chemometrics approach, the UVE-PLS model stands out with an R²Pred of 0.98 and an RMSEP of 1.41 for protein prediction, utilizing SNV and EMSC preprocessing. Meanwhile, for fat prediction, employing IPW-PLS with 15 variables and SNV resulted in an R²Pred of 0.94 and an RMSEP of 2.15. The best ML model for prediction of protein content had an R²Pred of 0.94 with a RMSEP of 2.14, which was based on preprocessing with EMSC. For fat prediction, the best performing model was also EMSC preprocessed, with an R²Pred of 0.97 and a RMSEP of 1.88.

Conclusion: We conclude that both methods have similar excellent performance when it comes to prediction of fat content and protein content in different cheese types. Notable is that ML achieved similar performance in a more naïve approach without variable selection. For the interpretation of which spectral bands contribute to the prediction outcome however, one cannot do without chemometrics. Together, these methods make a powerful toolkit in the analysis of hyperspectral images.

Control of high moisture extrusion texturization using pressure, torque and melt temperature

Clara Barnés-Calle¹, Pere Gou¹, Grau Matas¹, **Phd Elena Fulladosa¹**

¹Institute of Agrifood Research and Technology, Food Quality and Technology Program

Aim:

Protein texturization during high moisture extrusion process (HMEP) is influenced by multiple factors such as water feed, extrusion temperature and formulation (process inputs). At the same time, these factors determine the melt temperature, pressure and torque applied to the mixture (process outputs). The aim of this study was to explore the use of process outputs to predict product texture as a first step towards inline and real-time control of protein texturisation.

Method:

High-moisture extrudates (HME) were elaborated from different formulations of fava bean protein concentrate and its combination with oil and pea protein isolate using a laboratory-scale twin-screw extruder. All formulations were elaborated at different barrel temperatures (145 °C and 155 °C) and water feed rates (60% and 50%). During HMEP, melt temperature, pressure, and torque were recorded. Texture of all HME was analysed applying a shear test, both longitudinally and transversally to the extrusion flow, and the anisotropy index (AI) was calculated from the ratio between the transversal shear force (F_T) to the longitudinal shear force (F_L). Prediction of HME texture using process outputs as model variables was explored. Because collinearity between pressure and torque was identified, Principal Component Analysis (PCA) was also performed over the process outputs and the two principal components (PCs) were used to model HME texture.

Results:

Collinearity between pressure and torque compromised the prediction of HME texture when using process outputs as model variables. However, the use of PC1 (66.3% explained variance) and PC2 (30.6% explained variance) as model regression variables allowed the prediction of F_T , F_L , and AI with $R^2 = 0.77$, 0.72 and 0.58 (and RMSE = 3.383 N, 2.465 N·s and 0.1464), respectively. This indicates that although the resulting models do not allow a highly precise prediction of texture, monitoring of process outputs could be useful to control HME texturisation to an extent.

Conclusion:

Process outputs can provide useful information to predict HME texturisation and could be a first step to provide a real-time control of HME texture quality. However, the integration of this information with data collected from other non-invasive sensors (i.e.: near infrared) needs to be investigated to develop more precise control tools.

Real time quality assessment and process optimization of rice milling processes using artificial intelligence

Mr Benjamin Ilo¹, Dr Hongwei Zhang²

¹Sheffield Hallam University, ²National Centre of Excellence for Food Engineering

Aim

This research work is to develop an artificial intelligence (AI)-based automated real-time approach to evaluate the quality of rice products produced during the rice milling process by extracting milled rice physical features for quality analysis and provide additional data stream that can be used for real-time process optimisation as well as early-stage anomaly detection. Due to the flow volume of milled rice during milling processes, real-time image processing of rice milling frequently produces indistinct segmentation and classifications, which reduces the accuracy of the image analysis. However, real-time analysis can be accomplished by utilising AI, computer vision, and machine learning (ML) approaches.

Method

Computer vision and image processing techniques have had great success in the food and drink industry. These technologies are used to acquire real-time rice images; the acquired images will be converted to grayscale, labelled frames for processing and extracting numerical data from the images. AI techniques such as the Mask R-CNN and YoloV8 for instance segmentation are computer vision task used for object detection, image feature extraction, pixel-level segmentation, and grain classification. It assigns the same pixel values to all objects of the same class, capable of predicting bounding boxes and class probabilities directly from raw images in real-time. Numerical data generated are used to create real time closed loop for process optimization. This technique evaluates the real-time physical characteristics of the milled rice, such as the length, width, and colour.

Result

First-stage image processing, segmentation, and classification do not perform well under ideal operating conditions, but with AI and deep learning, real-time analysis results were satisfactory with 98% accuracy.

Conclusion

This research work will assist industries in evaluating the real-time physical characteristics of the milled rice, such as the length, width, and colour. It will be able to distinguish between good rice and broken rice and further provide additional data stream that can be used for real-time process optimisation as well as early-stage anomaly detection. This project is critical to running an optimal operation for the rice milling process. It will enhance the long-term viability of the milling process and boost its operational effectiveness.

Development of a probabilistic mycotoxin (DON) exposure assessment in flatbreads: a case study of Italy

Myrsini Kakagianni¹, Francesca Vurro², Antonella Pasqualone², Vasilis Valdramidis^{3,4}

¹University Of Thessaly, ²University of Bari, ³University of Malta, ⁴National Kapodistrian University of Athens

Aim: Flatbreads constitute a major food consumed worldwide. However, deoxynivalenol (DON) remains one of the most frequently mycotoxins produced by *Fusarium* species, due to the wide occurrence in highly consumed cereal-based food and its associated toxicological effects. The aim of this study was to assess the exposure of Italian population to DON related to the consumption of flatbread products through a probabilistic approach.

Method: The individual's body weight was taken into account and the consumption dataset was derived from Italian individual consumption data based on FAO with 97 occasions during the survey Italy-INRAN SCAI 2005-2006-CREA-Alimenti e Nutrizione covering the geographical regions North-West, North-East, Centre, South and Islands. The mean occurrence values, based on median DON concentrations, were 43.5 µg/day for unleavened bread, crisp bread and rusk. Consumption and occurrence data were both codified according to the FoodEx2 classification system developed by EFSA. The food products examined were: (i) Pizza base, cooked, INGRED=Vegetable fats and oils, edible, INGRED=Salt, (ii) Pizza base, cooked, process=Air heat drying and (iii) Unleavened or flat bread. Dietary exposure was assessed at individual level by multiplying the average daily consumption of each food with the mean DON occurrence summing up the respective intakes throughout the diet and finally dividing the results by the individual's body weight. Dietary exposure assessment was performed using ImproRisk model V0.5.4.

Results:

The distribution of exposure to DON, based on median occurrence scenario, showed that no percentage exceeds the tolerable daily intake (TDI=1 µg/kg bw/day) for the Italian population. It was also found that toddlers, other children and adolescents had the highest exposure values after consumption of two pizza bases. However, Hazard Quotient (HQ=Estimated Daily Intake-EDI) for these population groups was still less than 1, indicating the health risk of the intake level is acceptable.

Conclusion: The current results suggest the flatbread products marketed and produced in Italy present low risk for average and excessive consumers regarding the content of mycotoxin DON.

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Establishment of machine learning models for shear viscosity prediction of hydrocolloids over temperature

Seunghun Lee¹, Suyong Lee¹

¹Department of Food Science & Biotechnology and Carbohydrate Bioproduct Research Center, Sejong University

Aim :

Hydrocolloids are extensively used to control the rheological properties of various food products including viscosity. The measurement of their viscosity often relies on instrumental analysis as it is highly influenced by various factors such as concentrations and temperatures. Therefore, it is necessary to establish a framework for predicting hydrocolloid viscosity beyond the instrumental measurement.

Method :

The changes in the shear-viscosities of 8 hydrocolloids by temperature variations were measured using a controlled stress rheometer. Temperature variations were implemented through a heating and cooling cycle of 30°C-90°C-30°C. Machine learning models for predicting shear-viscosities of hydrocolloids over temperature were constructed in a python programming environment with a Jupyter Notebook.

Results :

Hydrocolloids exhibited different viscosity profiles during the heating and cooling cycle. Overall, except for methyl cellulose, most of the hydrocolloids exhibited a viscosity pattern where viscosity decreased with heating and increased with cooling. When the viscosities of the hydrocolloids were subjected to machine learning analysis, most of the machine learning models demonstrated excellent predictive performance by showing higher coefficients of determination and lower RMSE. However, their prediction performances varied depending on the type of machine learning models.

Conclusion :

In this study, the viscosities of 8 hydrocolloids over temperature experimentally measured were successfully predicted by machine learning models ($R^2 > 0.9$). The results of this study suggest that machine learning can be utilized as a new predictive framework for predicting the rheological properties of hydrocolloids in response to temperature changes.

Enhancing precision nutrition strategies to favour healthy aging through Neural Networks-based algorithms development

PhD Adrián Martín-Segura¹, PhD Laura J. Marcos-Zambrano¹, Blanca Lacruz-Pleguezuelos¹, PhD Alberto Díaz-Ruiz¹, PhD Enrique Carrillo de Santa Pau¹

¹IMDEA Food

Aim:

Aging is the greatest risk factor for the development of chronic diseases including cardiovascular disorders or cancer. Since the rise in life expectancy puts a high pressure on healthcare systems, mechanisms to promote healthy aging, defined as aging free of disease, through non-pharmacological means needs to be identified. Nutrition, seen as a powerful medicine to influence human physiology, has demonstrated effectiveness at slowing aging progression via different processes, including metabolism and/or lowering inflammation. Likewise, the gut microbiome which comprises all gut microorganisms, is strongly influenced by nutrition and has shown to modulate these aging-related mechanisms. Thus, studying gut microbiome architectures along aging could help us to develop and evaluate nutritional interventions to modulate these communities, strengthening healthy aging.

Method:

We are characterizing gut microbiome changes along aging, considering also alterations on microbiome derived metabolites due to these changes. To do so, we are using neural networks (NN), advanced artificial intelligence algorithms with great capability to integrate complex data. We have already identified ~3700 whole genome shotgun samples from public repositories associated with healthy aging to train these algorithms, that will integrate microbiome and metabolite data and allow us to identify patterns on microbiome that are associated with age, sex or geography, unveiling aging biomarkers we could use to design nutritional interventions.

Results:

Microbiome α -diversities and taxonomies along age differ when adjusted by sex, geography or BMI. Interestingly, the highest α -diversity (~30% more) is found in industrialized countries, with a strong presence of *genus Akkermansia* only in developed countries' elders. Also, for BMI<30 we observe high values of α -diversity among 80 years or older subjects. We have tested different NN architectures (e.g.: convolutional, GraphNN) integrating taxonomic abundances and metabolite signatures, observing trends in these metrics that are showing new microbiome patterns related to aging.

Conclusion:

We have developed different NN architectures to identify new microbiome biomarkers associated with aging. Due to their integration power, these algorithms reveal as a great asset to design and evaluate nutritional interventions efficacy on subjects' health, enhancing the identification of new mechanisms to favour healthy longevity.

Unraveling Key Parameters for Enzyme-Assisted Bioactive Compounds Extraction from Agrifood Residues: A Machine Learning-Driven Meta-Analysis

Fatemeh Mojarradi¹, Francesco Donsi¹, Alessandra Procentese¹

¹University Of Salerno

Aim:

Nowadays, enzyme-assisted extraction (EAE) has been used in research as an innovative alternative to traditional methods for bioactive compounds extraction. This approach is gaining popularity due to its effectiveness and environmental benefits. The EAE process is influenced by several parameters, such as temperature, enzyme concentration, time, pH, solvent-to-sample ratio, the nature of the target compounds, and the specific enzyme used for extraction. Analyzing the impact and importance of each parameter is essential for optimizing the extraction process and, consequently, managing the financial implications. This study is a machine learning-driven meta-analysis that unravels the key parameters of EAE for bioactive compound extraction from agrifood residues.

Method:

The meta-analysis of studies was conducted using two machine learning models, random forest and gradient boosting, developed from data extracted from literature articles. The accuracy and performance of these models were assessed using root-mean-square error (RMSE) and mean absolute error (MAE).

Results:

The results indicated that the random forest and gradient boosting models significantly outperformed the multilinear regression model, which was used as a benchmark. Furthermore, these machine learning techniques have proven to be reliable methods for analyzing and identifying the key parameters influencing the EAE process. Based on the results, pH and enzyme concentration were identified as the most important parameters in this process.

Conclusion:

This study shows the machine learning models as reliable tools for meta-analysis of the most important parameters of the EAE process.

Towards autonomous bioprocess control: Monitoring, Predicting and Controlling pilot scale outdoor Photobioreactors for microalgae cultivation.

Eric Morelle¹, Cornelia Rauh¹

¹TU Berlin

Aim:

To develop a predictive framework for optimizing microalgae cultivation in pilot-scale outdoor photobioreactors, where weather conditions have a significant impact on algae growth. The goal is to create an autonomous control system that adapts to changing environmental conditions and optimizes photobioreactor operation without necessarily relying on costly and energy-intensive light and temperature control systems.

Method:

We monitor the growth of microalgae in two 100L photobioreactors operated in a greenhouse and exposed to varying light and temperature conditions. Online monitoring of growth parameters such as pH, temperature, and biomass concentration provides real-time insights into the bioprocess. Weather data from nearby meteorological stations is collected to provide a reference for the prevailing weather conditions. The collected data is used to train machine learning models that predict the changes in process variables with respect to their state and the environmental conditions. Predictions are then made by incorporating the models and weather forecasts into the construction and solution of a classical initial value problem.

Results:

Our predictive models accurately predict algae growth patterns, enabling the development of control strategies to maximize yields. The combination of machine learning models and ordinary differential equations allows for rapid adaption of process dynamics and the prediction with arbitrary time step lengths. We show that the concept is applicable to various biological and physical quantities. By integrating real-time monitoring and continuous model retraining, the predictive models reflect the most recent state of process observations.

Conclusion:

The approach demonstrates the potential of combining process monitoring and machine learning for the prediction and control of photobioreactors to optimize outdoor microalgae cultivation by adapting to environmental conditions. Since the applied principles are not limited to microalgae processes, applications in other environment-dependent bioprocesses like farming or aquaculture are imaginable. The developed framework will be used as a foundation for more advanced control strategies, such as reinforcement learning, to improve bioprocess performance.

Agile methodologies and software used to design healthy and sustainable food in SMEs Latin America

Jairo Alonso Torres Garcia¹, Letricia Barbosa¹, Joao Lima^{2,3}

¹FoodChemPack Research Group, Department of Analytical Chemistry, Nutrition and Food Science, Faculty of Pharmacy, University of Santiago de Compostela, Campus Vida, 15782 Santiago de Compostela, Spain, , ²H&TRC - Health & Technology Research Center, Coimbra Health School, Polytechnic University of Coimbra, Coimbra, Portugal., ³GreenUPorto - Sustainable Agrifood Production Research Centre, Porto, Portugal.

Introduction:

The design and formulation of food, an intricate and prolonged process, faces challenges in SMEs, where it often relies on trial and error, limiting its competitiveness in the market. Given the need for agile and sustainable solutions, this research addresses the current use of agile methodologies in the development of food products in SMEs.

Objective:

To identify agile methodologies and software used to design healthy and sustainable food in SMEs in Latin America.

Method:

An ethnographic study was conducted in 5 Latin American countries (Brazil, Colombia, Chile, Mexico, and Uruguay), through semi-structured interviews with 14 food design managers, based on a questionnaire of 6 questions with a maximum duration of 30 minutes. Additionally, the AEIOU non-participant observation method was applied through on-site visits to SMEs or analysis of visual materials.

Results:

The sample included 11 food engineers, 1 chef, 1 electronic engineer, and 1 nutritionist, belonging to 2 multinational companies, 4 medians to large enterprises, and 8 SMEs, covering food additives, dairy, meat, soups, and various products. The majority indicated that the food design process takes between 1 and 2 years approximately. The main limitations identified were lack of raw material availability, high production costs, limited knowledge of regulations and lack of clear guidelines, expensive processes, cultural barriers, and the need for production for subsistence. Regarding knowledge of agile methodologies, SMEs indicated a lack of knowledge about methodologies and software; medium-sized companies only mentioned the use of the Stage-Gate method; while large companies reported having internal standardized methods, without revealing details due to corporate policies. In the non-participant observation, it was found that SMEs lack sophisticated equipment and software assistance for food design.

Conclusion:

Latin American SMEs show limited use of agile methodologies and software to design healthy and sustainable food.

Understanding Shifting Insect Pest Behavior and Disease Dynamics under Climate Change: Rule-Based Machine Learning Approach

xinxin wang¹, Ali Hürriyetoglu¹, Cheng Lin¹, Samuel Sutanto¹, Phd Bas van der Velden

¹Wageningen Food Safety Research

Aim:

Climate change significantly impacts agricultural systems, leading to shifts in insect pest behavior and disease dynamics. This study analyses shifts of insect pests and disease occurrences under climate change.

Material and methods:

Through rule-based machine learning, we analyzed the relationship between insect pest occurrences and weather variables (including daily air temperature, dewpoint, relative humidity, precipitation, and wind speed) using data obtained from apple fields collected between 2020-2023. Data included direct farmer observations (observed pest occurs: positive or negative), expert-developed pest alarms, and weather data. The data are sparse and not systematically collected, including 45 (7%) positive records, 320 (50%) negative records, and 277 (43%) unlabeled records (i.e., labeled 'uncertain'). We employed K-means clustering for cluster pattern analysis. We applied several tree based models (decision tree, random forest, and XGBoost) for predictive modeling of pest and disease occurrence in apples. These tree based models were used to reflect associations between weather variables and pest and disease occurrences. Data were separated into a training dataset (80%) with 5 fold cross validation, and a hold-out testing dataset (20%). Model performance was evaluated using accuracy, precision, recall, F1, and confusion matrix.

Results:

K means clustering showed distinct clusters of pest occurrence associated with specific weather conditions, with higher risk periods identified during certain months characterized by low wind speed, relative humidity, precipitation, and high temperatures. Such rules derived from data were confirmed by domain experts to be highly relevant to pest and disease occurrence in apples. Among the tree based models, XGBoost showed the highest model performance with mean accuracy of 0.98, F1 score of 0.94, recall of 1, precision of 0.88 on the training dataset, and accuracy of 0.82, F1 of 0.35, recall of 0.42, precision of 0.32 on the testing dataset.

Conclusion:

Climate variables were associated with insect pests and disease occurrences. The predictive model did not generalize to the hold-out testing data set. This is likely due to the limited amount of data at the moment. We are currently systematically collecting insect pest and disease observations to improve the generalization of the predictive model.

"Optimizing Food Machinery: Utilizing LSTM Anomaly Detection for Predictive Maintenance"

Mr Syed Haseeb Haider Zaidi¹, Alex Shenfield, Hongwei Zhang

¹Sheffield Hallam University

Aim:

Reliability and efficiency of machinery are vital for ensuring product quality, safety, and operational continuity. Unexpected equipment failures can lead to costly downtime and compromise food safety standards. Proactive maintenance strategies, facilitated by anomaly detection techniques, offer a promising approach to identify potential issues before they escalate. This study focuses on utilizing Long Short-Term Memory (LSTM) models for anomaly detection in food machinery, aiming to enhance equipment reliability and minimize operational disruptions.

Method:

The experiment employs motor current data to detect anomalies indicative of damaged bearing states in food machinery. The dataset comprises instances of both damaged and non-damaged bearing states, providing a realistic representation of operational conditions. Data preprocessing involves normalization and windowing techniques to facilitate model training. An LSTM model architecture, featuring a single LSTM layer followed by a dense layer for prediction, is trained using the Adam optimizer and mean squared error loss function.

Results:

The LSTM-based anomaly detection model demonstrates robust performance, achieving an overall Root Mean Square Error (RMSE) of 0.5 on the test dataset. Anomalies are detected based on prediction errors exceeding predefined thresholds, highlighting deviations from expected motor current patterns. Visual analysis of the anomalies detected during the evaluation process provides valuable insights into potential equipment malfunctions, facilitating proactive maintenance interventions.

Conclusion:

This study underscores the importance of anomaly detection in food machinery maintenance and safety. By leveraging LSTM models for anomaly detection, food processing facilities can implement proactive maintenance strategies that enhance equipment reliability, minimize downtime, and ensure product quality and safety standards. These findings contribute to the advancement of maintenance practices in the food processing industry, ultimately supporting operational efficiency and consumer trust.

Building the future food system with DevOps methodology

Ms Jiaqi Zhou¹, Prof Paul Brereton¹, Prof Katrina Campbell¹

¹Queen's University Belfast

Aim:

The DevOps methodology has traditionally been associated with computer science, but is now being used in non-traditional areas such as food systems. This transition is driven by the recognition that DevOps principles—agility, responsiveness, and continuous improvement—can significantly enhance the performance of intelligent food systems.

Method:

The study utilizes a comprehensive review of the current literature, case studies, and existing technology frameworks to identify critical DevOps strategies. These strategies are aimed at improving the development of food systems, covering all stages from planning to operation.

Results:

The findings of the study suggest that the identified potential technologies may contribute to the development of future intelligent food systems. The review indicates that, although DevOps currently lacks the use of cases in the food sector, it demonstrates significant potential for growth.

Conclusion:

This review serves as a technical guide for future food systems and can also provide a foundational process for enabling intelligent food systems. The paper concludes that the DevOps methodology, with its emphasis on cross-functional collaboration and automation, can play a crucial role in driving innovation within food systems in the future.

Identification of emerging risks in the food chain with advanced topic detection methods

Dr Zsuzsa Farkas¹, Krisztián Vribék¹, Szilveszter Csorba¹, Dr Ákos Józwiak¹

¹University of Veterinary Medicine, Digital Food Institute

Aim: Ensuring the resilience of food systems necessitates a systematic evaluation of trends and emerging risks across short, medium, and long timescales. However, there are several factors that make this process a complex, interdisciplinary task. Risks may arise from different emerging hazards, which are well defined, yet to be examined, however, complex, driver-induced early signals and the increase of exposure for known hazards also must be considered.

Discerning developing patterns in the food chain is essential for strategic planning and analysis, facilitating informed decision-making processes. The early recognition of emerging risks not only safeguards human, animal, and plant health but also plays a crucial role in surveillance planning. Additionally, it provides valuable input for the formulation of risk management, mitigation, and prevention measures.

Method: The identification process is facilitated by data and text mining algorithms such as topic detection. Topic detection involves the identification and extraction of prevalent themes or subjects within a body of text. It analyzes textual data to discern key topics, enabling researchers and businesses to gain insights into trends, or sentiments within a given set of documents. It is particularly valuable for understanding the prevailing themes in large datasets. The food safety news from Europe Media Monitor has been used as an input data for emerging risk identification.

Results: The results of topic detection methods and the identified trends and emerging risks such as sustainable packaging solutions, essential oils and nanoplastics for antimicrobial use, e.g. micro- and nanoplastics, migration from food contact materials; will be presented in the lecture.

Conclusion: The timely detection of emerging signals and risks offers the opportunity to implement essential risk mitigation measures, thereby preventing the escalation of potential risks. Adapting food systems to current and evolving trends in innovation, technology, and consumer behaviors is pivotal for success and sustainability.

Experimental Analysis and Predictive Modeling using Machine Learning of Spontaneous Emulsification

Fatemeh Mojarradi¹, Francesco Donsi¹

¹University Of Salerno

Aim:

Emulsions play a key role in the food industry, contributing to enhancing the stability and functionality of various products. Whether in the food, cosmetics, or pharmaceutical industries, the capability to produce emulsions with controlled size distribution and high stability is of paramount importance to product quality, ensuring optimal performance and consumer satisfaction. Spontaneous emulsification is based on the spontaneous (or “catastrophic”) emulsification of an oil phase, where a hydrophilic surfactant has been dispersed when mixed with water. The process of spontaneous emulsification is influenced by diverse parameters, including oil concentration, surfactant-to-oil ratio (SOR), oil density, interfacial tension, viscosity, surface tension, pH, temperature, hydrophilic-lipophilic balance (HLB), molecular weight of oil, molecular weight of surfactant, and critical micelle concentration (CMC), which are all relevant in controlling the emulsion droplet size. This study focuses on the application of machine learning in constructing predictive tools for the process of spontaneous emulsification based on the random forest and gradient boosting models.

Method:

The machine learning models were developed based on literature data extracted from articles and experimental data obtained from a series of experiments conducted under different spontaneous emulsification process conditions using surfactants such as Tween 20, Tween 40, Tween 80, soy lecithin, and monoolein, and the oil phase included medium-chain triglycerides (MCT), sunflower oil, coconut oil, and Captex 300. The performance and accuracy of the models were evaluated through root-mean-square error (RMSE), mean absolute percentage error (MAPE), mean absolute error (MAE) and coefficient of determination (R^2) using a different dataset from the one used for training.

Results:

The results showed that, in comparison with a multilinear regression model, used as benchmark, both random forest and gradient boosting models had a significantly better predictive performance. RMSE reduced from 0.5359 to 0.3042 and 0.2573 in random forest and gradient boosting, respectively. Additionally, the obtained R^2 for both random forest and gradient boosting were higher in comparison with multilinear regression model (0.3194). **Conclusion:** The study demonstrates the feasibility and effectiveness of using machine learning models as predictive tools for the spontaneous emulsification process for the formation of O/W emulsions with the desired droplet size.

Advanced Meat Processing Utilizing Statistical Shape Modeling for Internal Bone Prediction in Pork Shoulders

Advanced Meat Processing Using Statistical Shape Modeling For Internal Bone Prediction In Pork Shoulders Michiel Pieters¹, Pieter Verboven¹, Bart Nicolai^{1,2}

¹Postharvest group, Division MeBioS, Department of Biosystems, KU Leuven, Willem de Croylaan 42, 3001 Leuven, Belgium, ²Flanders Centre of Postharvest Technology, Willem de Croylaan 42, 3001 Leuven, Belgium

Aim:

Deboning of meat is a labor-intensive process that is still performed manually. There is an interest in advanced automation of the processing chain to replace or assist the deboning operations. Thereof, inline inspection systems are explored that can analyze the bone structure of pork shoulders. A computed tomography (CT) system would be suitable as the X-rays can penetrate the large volumes of meat resulting in high-contrast reconstructed 3D images of the meat and bone. This 3D representation of a pork shoulder can be fed to an automation system. However, CT systems have a few disadvantages like its size and cost. Using a statistical shape model, prior knowledge of the pork shoulder can be used, this makes it possible to estimate the inside 3D bone structure while only measuring the outside shape of the meat sample with a 3D camera.

Method:

This research focuses on the development of a statistical shape model (SSM) specifically tailored to characterize the relationship and variability between the external shape of pork shoulders and their internal bone structure. To build this model, we collected a dataset of 90 CT measurements of pork shoulders. These measurements were obtained using high-resolution CT systems at the KU Leuven XCT core facility and a gantry CT system at UZ Leuven. Utilizing this dataset, we created reference 3D models of both the outer shape and bone structure of the pork shoulders.

Results:

The developed statistical shape model accurately estimates the internal bone structure solely from the externally measured shape using a 3D camera. 80 pork shoulder samples were utilized for model development, with 10 additional samples reserved for validation. The worst-performing fit in the test set had only a mean average error (MAE) of 6.93 mm.

Conclusion:

The developed statistical shape model (SSM) demonstrates promising results in predicting the inside bone structure of pork shoulders from their external shape. However, further research is necessary to assess the model's robustness. Moreover, this combined SSM can generate realistic synthetic 3D models of pork shoulders, presenting opportunities for various applications in this field.

Hydrothermal treatments derived ensemble machine learning models: Safety assurance of *Phyllospora comosa* biomass

Dr Thiruchenduran Somasundaram¹, Dr Thomas Mock¹, Dr Damien Callahan², Dr David Francis¹

¹Nutrition and Seafood Laboratory (NuSea.Lab), School of Life and Environmental Sciences, Deakin University, ²School of Life and Environmental Sciences, Deakin University, Burwood Campus

Aim: This study aims to employ hydrothermal experimental data to train machine learning models to develop, optimise and dictate hydrothermal treatments to reduce iodine and arsenic below their Australian regulatory levels.

Method: This study employed a model hydrothermal experiment utilising varying temperatures (22 °C, 52 °C, and 82 °C) and exposure times (control = 0, and 2, 10, 50, and 250 seconds) to dictate the time necessary to reduce the iodine and arsenic concentrations of *Phyllospora comosa* below their Australian maximum residual limits (iodine = 1 mg/g and arsenic = 0.00667 mg/g; dry weight).

Results: The treatments reduced iodine from 2.76 mg/g to 0.88 mg/g, and arsenic concentration from 0.01693 mg/g to 0.00965 mg/g, respectively. The ensemble predictive machine learning models showed that a treatment at 100 °C for 3 minutes and 58 s would reduce the arsenic concentration to 0.00667 mg/g. In addition, a treatment at 100 °C for 10 minutes and 41 s would reduce the iodine concentration to 0.02 mg/g, equivalent to the iodine concentration of commercial nori sold in Australia.

Conclusion: Hydrothermal treatments have the potential to produce safe and nutritionally enhanced seaweed products. Further, it is proposed that a hydrothermal treatment at 100 °C for 4 minutes will ensure the safety of similar seaweed biomasses. The methods and the models described herein can be adopted in industrial algal food processing with warranted experimental validations.

Modelling and prediction of quality attributes in yogurt-like analogues using chickpea, oat and insect proteins

Simone Villiot^{1,2}, Dr Saioa Alvarez-Sabatel¹, Patricia Rioja¹, Laura Fernández-Lucio¹, **Dr Clara Talens¹**
¹AZTI, Food Research, Basque Research and Technology Alliance (BRTA), ²Department of Food and Drug, University of Parma, Italy

Aim

The aim of the study was to develop predictive models for the formulation of protein-enriched yogurt-like analogues, using a desirability-based mixture design model. Two extensive datasets were analysed for ingredient frequency and nutritional profiles, and multiple linear regression was employed to optimize the texture, colour, and protein content to attain a "source of protein" claim.

Method

Desk research using two datasets initially assessed the frequency and characteristics of ingredients in yogurt-like analogues. The first dataset, sourced from Google Scholar, included 222 pages of results from 2023, from which 11 articles and 2 patents were reviewed, covering 71 formulas for yogurt-like analogues. The second dataset analysed the ingredient list of 1787 commercial yogurt-like analogues from the Mintel database. Control samples (commercial) were selected based on textural properties, flavour, nutritional value, and ingredients. The experimental design was targeted to ensure that at least 12% of the total caloric value was derived from proteins. A desirability-based mixture design model considered 7 combinations of chickpea protein, oat protein and hydrolysed insect protein. Multiple linear regression predicted protein content (Kjeldahl), instrumental colour ($L^*a^*b^*$), and texture (firmness, consistency, cohesiveness, and viscosity index). The optimized top 3 samples (highest desirability) were evaluated via Quantitative Descriptive Analysis (QDA), with assessors re-trained for specific texture and flavour profiles.

Results

The predictive models effectively assessed the quality attributes of yogurt-like analogues with an R^2 value from 0.89 to 0.98, indicating a high level of predictive accuracy. Instrumental texture analysis showed analogues with a higher hydrolysed insect protein ratio exhibited firmness and viscosity indices approximating traditional yogurt. Colorimetric analysis demonstrated chickpea protein-based analogues had higher L^* values, indicating a lighter colour, while oat protein variants displayed a beige tint. Protein analysis revealed chickpea protein analogues possessed the highest percentage of caloric value derived from proteins (12.5%). Sensory evaluations favoured oat and chickpea variants for their smooth texture and mild flavour, suggesting strong consumer acceptance potential.

Conclusion

By applying robust statistical and modelling techniques, this study anticipates establishing a foundational understanding of how non-traditional protein sources can be effectively utilized in yogurt-like products, emphasizing sustainable alternatives in the food industry.

Innovative use of vacuum and ultrasound-assisted impregnation for enhanced functionality of Gala apple

M.Sc Marcellus Arnold¹, Dr Urszula Tylewicz², Professor Joanna Suliburska¹, Professor Michał Świeca³, Professor Anna Gramza-Michałowska¹

¹Poznań University of Life Sciences, ²University of Bologna, ³University of Life Sciences in Lublin

To achieve goal number 2 and 3 of Sustainable Development Goals (achieving food security, ensuring healthy lives), the development of functional foods based on local commodities is encouraged. Apples and sea buckthorn (SB) grow well in Asia and Europe; they provide health-promoting effects based on recent studies. In preliminary study, freeze-dried Gala apples conventionally impregnated with 93.8% (w/w in water) SB juice with 4% calcium lactate (CaL) at 30°C for 120 min was observed to have the optimum calcium content with minimum antioxidant activity loss. The application of non-conventional technologies during impregnation could further increase the calcium content and antioxidant activities of the product, which potentially prevents osteoporosis and calcium deficiency.

Aim:

This research aims to evaluate the impact of vacuum (VI) and ultrasound-assisted impregnation (US) on antioxidant activities, calcium content, and polyphenol-oxidase activities of freeze-dried Gala apple impregnated with 93.8% SB juice, with 0 and 4% CaL, compared to conventional impregnation (CON).

Method:

CON was conducted following the preliminary study with 0 and 4% CaL. VI (200, 400, and 600 mbar; 30°C) was applied for 10 min of holding and relaxation time. US (25 kHz, 1000 W, 30°C) was applied for 10, 20, and 30 min followed by CON for 110, 100, and 90 min, respectively. Antioxidant activities (ABTS, DPPH, FRAP, ORAC, photochemiluminescence/PCL), calcium content, and polyphenol-oxidase activity of freeze-dried samples were analyzed.

Results:

Samples prepared at 4% CaL solution showed higher calcium content but lower antioxidant activities than those at 0% CaL. The highest antioxidant activities were observed after VI at 200 mbar or US for 30 min at 0% CaL solution. The highest calcium content was observed after VI at 200 mbar (1.12 times higher than CON) or US for 10-30 min (about 1.68 times higher than CON) at 4% CaL solution. Polyphenol-oxidase activity was completely inhibited by CON and US for 30 min at 0% CaL.

Conclusion:

Compared to CON, VI and US using SB juice with 0 or 4% CaL improved the functional properties of freeze-dried apples. This study could provide a new alternative to healthy snacks that potentially prevent non-communicable diseases, especially osteoporosis.

Whey as a source of lactoferrin

Dr. Klára Bartáková¹, Prof. Lenka Vorlová¹, Prof. Oto Hanuš², Hana Nejeschlebová², Dr. Pavlína Navrátilová¹, Dr. Sandra Dluhošová¹

¹University of Veterinary Sciences, ²Dairy Research Institute Ltd.

Aim:

Lactoferrin is among the natural inhibitory substances of milk. Its important functions include antibacterial and antiviral activity, which have been proven by a number of studies for both human and animal populations. Other abilities include antifungal and antiparasitic action, as well as support for the proliferation, differentiation and activation of cells of the immune system and the strengthening of the immune response.

From the point of view of its chemical nature, lactoferrin is a whey protein, therefore dairy products made from concentrated whey, together with other biologically active substances, can significantly affect the health of consumers. The aim of this study is to search for parameters that influence the content of lactoferrin in milk.

Method:

Cow's milk samples were taken from farms in the Czech Republic a total of four times, 30 samples each at the end of autumn, winter, spring and summer during the autumn 2022 to summer 2023 season. Milk was defatted by fat centrifugation and then precipitated with acetic acid to pH 4.6. The proportion of lactoferrin in the centrifuged whey was determined by means of a liquid chromatograph with a photodiode array detector.

Results:

The highest lactoferrin content of 209 ± 126 mg/l was determined in milk at the end of the winter period, while the lowest amount of 122 ± 116 mg/l at the end of the autumn period. The differences are not statistically significant. The greatest variability of the results was found in the spring period (176 ± 166 mg/l). Some authors state that the content of lactoferrin depends on the order of lactation, but we found a statistically significant ($p < 0.05$) dependence only in the spring period. In contrast, increasing lactose content statistically significantly reduced the amount of lactoferrin in milk in all periods. Furthermore, a statistically significant dependence of the lactoferrin content on the total protein content was found.

Conclusion:

The above results show that the influence of individual factors is combined, therefore it is necessary to continue studying the content of lactoferrin in milk.

Acknowledgment:

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Global sensitivity analysis of the liver glucose metabolism model kinetic parameters

Miss Antonija Matos¹, Associate professor Davor Valinger¹, Assistant professor Tamara Jurina¹, Mrs Tea Sokač Cvetnić¹, **Associate Professor Maja Benković¹**, Professor Jasenka Gajdoš Kljusurić¹, Associate professor Ana Jurinjak Tušek¹

¹University Of Zagreb, Faculty Of Food Technology And Biotechnology

Aim: This study aimed to assess the collective effect of uncertain input parameters on the variability of predictions in a mathematical model of human carbohydrate metabolism in liver cells, utilizing a global parameter sensitivity analysis approach.

Method: The mathematical model utilized in this study was initially proposed by Kurata (2021) and consisted of 336 kinetic parameters, 80 variables, and 81 biochemical reactions. Differential equations were solved using WR Mathematica for various initial concentrations of glucose based on predefined scenarios, namely ketogenic, Mediterranean, and high carbohydrate diets. Given the inherent complexity and non-linearity of metabolic models, sensitivity analysis is crucial for validating parameter values. Specifically, the Fourier Amplitude Sensitivity Test (FAST) was employed, involving a 3% increase in each of the 336 kinetic parameters. This analysis focused on assessing the impact of parameter variations on glucose concentration in the blood and liver, glucose-6-phosphate concentration, and the rate of the reaction catalyzed by glucokinase.

Results: The findings indicate that steady-state concentrations of glucose in the blood and liver, as well as glucose-6-phosphate, are highly sensitive to changes in the saturation constant and maximum speed of the glucose transport reaction from blood to liver cells. Additionally, the rate of the reaction catalyzed by glucokinase is predominantly influenced by the saturation constant of pyruvate kinase for ADP in the liver.

Conclusion: This study underscores the importance of addressing input parameter uncertainty in metabolic modeling, as highlighted by the comprehensive sensitivity analysis conducted. Such global parametric sensitivity analyses offer valuable insights into critical nodes within the metabolic pathways.

Exploring the potential of Tuscan bee pollen: antioxidant activities and anti-inflammatory effects on pulmonary cells

Mr Andrea Cavallero¹, Francesca Vidotto¹, Carmen Lamacchia¹, Morena Gabriele¹

¹Institute of Agricultural Biology and Biotechnology

Aim: Among apicultural products, bee pollen represents one of the most complete natural foods and a great source of energy for human nutrition. Several studies pointed out good bee pollen therapeutic properties, including antioxidant, anti-inflammatory, antimicrobial, anti-mutagenic, and antitumor effects. Chronic obstructive pulmonary disease (COPD) is a progressive respiratory disease characterized by an irreversible obstruction of the airways, caused by both genetic and environmental factors, primarily cigarette smoking. COPD is associated with oxidative stress and chronic inflammation of the lung tissue. Bee pollen may hold promise for treating the inflammatory component associated with COPD. The present work aimed to characterize a Tuscan polyfloral organic bee pollen by evaluating its phytochemical profile and *in vitro* antioxidant activities. Finally, the anti-inflammatory effect of bee pollen extract was evaluated on a tumor necrosis factor-alpha (TNF- α) inflamed human pulmonary cell line (A549).

Method: The phytochemical profile of bee pollen was evaluated by detecting bioactive compounds such as polyphenols, flavonoids, and carotenoids. Additionally, antioxidant activities were determined by DPPH, FRAP, and ORAC assays. The gene expression of IL-8 and COX2 inflammatory markers was analyzed by Real-Time RT-PCR and immunoblot analysis in A549 exposed for 24 hours to TNF- α , after 1 hour of pre-treatment with bee pollen extract.

Results: Bee pollen's phytochemical profile revealed high concentrations of total polyphenols (8.40 ± 0.16 mg GAE/g fresh weight, FW), flavonoids (5.29 ± 0.4 mg QE/g FW), and carotenoids (10.41 ± 1.38 μ g carotenoids/g FW), accompanied by good antioxidant activity, as indicated by DPPH (89.2 ± 6.55 % ARA), FRAP (83188 ± 1475 μ M Fe²⁺), and ORAC (433.77 ± 18.95 μ mol TE/g FW) assays. Pre-treatment with bee pollen extract (10 and 50 μ g/ml) notably suppressed the expression of IL-8 and COX2 inflammatory markers in TNF- α -stimulated A549 cells, demonstrating substantial anti-inflammatory effects. Western blot analysis further illustrated reductions in COX2 protein levels and attenuation of NF- κ B signaling activation.

Conclusion: In summary, bee pollen extract emerges as a promising nutraceutical product with significant potential for both medical and nutritional applications. Its demonstrated efficacy in mitigating inflammation suggests its utility in preventing a range of diseases associated with this process, including COPD.

Extraction of anti-inflammatory fractions from *Agaricus blazei* using supercritical CO₂

Paula García Ponsoda¹, Laura Jaime¹, Alejandro Ruiz-Rodríguez¹, **Rebeca Lavega**, Cristina Soler-Rivas¹, Susana Santoyo¹, María Trinidad Herrero²

¹Facultad de Ciencias. Universidad Autónoma de Madrid, ²Facultad de Medicina. Universidad de Murcia

Aim:

Agaricus blazei is considered as a medicinal mushroom by traditional Asiatic medicine because it has been used to prevent disorders such as atherosclerosis, hepatitis, hyperlipidemia, diabetes, dementia, dermatitis, cancer etc. Recently, scientific studies confirmed that they showed natural compounds with health benefits although their mechanism of actions are still unknown. Some of those bioactive compounds were low polar molecules as fungal sterols and derivatives and therefore, they could be concentrated with SFE (supercritical fluid extraction) technologies. Thus, *A. blazei* fruiting bodies were submitted to SFE extraction to obtain fractions and their anti-inflammatory activities were evaluated.

Method:

The SFE extractions were carried out at 300 bar, 40 °C during 3h with or without ethanol as co-solvent. The anti-inflammatory activity was determined using human monocytes THP-1 differentiated to macrophages (THP-1/M). The release of pro-inflammatory cytokines TNF- α , IL-1 β and IL-6 was measured in THP-1/M supernatants. Fungal sterols were identified by GC-MS.

Results:

SFE extractions from *A. blazei* showed very low yield suggesting a high sterol selectivity. The extracts reduced secretion of pro-inflammatory cytokines (TNF- α , IL-1 β and IL-6) when applied at low concentrations (15 μ g/mL) to differentiated macrophages activated with LPS. Extracts obtained with pure supercritical CO₂ showed higher inhibition of the cytokines secretion than with co-solvent. This anti-inflammatory activity seemed to be related with the presence of fungal sterols, such as ergosterol and ergosterol peroxide contents in extracts, since both sterols were effective against THP-1/M cells inflammation. Further studies are being carried out at the present to investigate the influence or synergies with other compounds from the extracts.

Conclusion:

These results pointed SFE extraction as a suitable method for obtaining fungal sterols-enriched extracts with anti-inflammatory activity. Particularly, extracts obtained without co-solvent.

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Mineral profile of wild edible mushrooms from Sierra Norte in Puebla MÉXICO

Dr Diana Guerra-ramírez¹, Dr Irma Salgado-Escobar², MC Tonathiu Rescendiz-Gutiérrez³

¹Universidad Autónoma Chapingo, ²Tecnológico de Monterrey, ³Universidad Autónoma Chapingo

Aim: The mushrooms have the capacity of absorbing elements from the different types of soil where they develop. It is important to especially analyze the type of element in edible mushrooms to determine if they are beneficial or toxic. The objective of this work was to complement the traditional knowledge and provide security guidelines for the wild edible mushrooms consumption in the Zaragoza and Zacapoaxtla, Puebla region.

Method: Seven work species were selected: *Amanita jacksonii*, *Helvella lacunosa*, *Hypomyces lactifluorum*, *Imperator torosus*, *Infundibulicybe squamulosa*, *Lactarius indigo* and *Lactifluus gerardii*, these were collected in the traditional markets in Zaragoza and Zacapoaxtla, Puebla. The different types of elements were analyzed by inductively coupled plasma mass spectrometry (ICP-MS). Particularly, it was quantified the concentration of 14 elements (Mn, Fe, Cu, Zn, V, Cr, Ni, Se, Sr, Ag, Hg, As, Pb and Cd) and their Health Risk Index.

Results: Of the elements analyzed, 20 were selected and classified into three groups: macroelements (Na, Mg, Al, Ca, Mn, Fe, Cu and Zn), trace elements (Li, V, Cr, Co, Ni, Se, Sr, Ag and Hg) and toxic elements (As, Cd and Pb). The species *H. lactifluorum* presented high concentrations of macroelements (Al, Ca, Mn and Fe), trace elements (Li, V, Co and Sr) and toxic elements (As and Pb). Likewise, *L. gerardii* had high concentrations of Pb and Cd. The health risk index (HRI) of the fungi under study is at safe levels (< 1) for Mn, Zn, V, Ni, Se, Sr and Pb in all species, however, high concentrations were found of Hg. Although the species *I. squamulosa* and *H. lactifluorum* have a high HRI, they do not constitute a risk because they are consumed in limited quantities and once a year.

Conclusion: Based on the results, it was found that seven elements (Fe, Cu, Cr, Ag, Hg, As and Cd) exceed security limits. Although the wild edible mushroom consumption is seasonal, it is recommended to ingest amounts that do not represent a health risk.

Use of Germinated Mung Bean (*Vigna radiata*) Flour in Beef Meatballs

Aybike Kamiloğlu¹, Tuğba Elbir Abca¹, Ahmet Akköse²

¹Bayburt University, ²Atatürk University

Aim: Germination is a method used for centuries to increase the amount of nutritional components, reduce antinutritional properties and soften the grain grain. Decomposition of high molecular weight polymers during germination leads to the production and development of biofunctional substances. When considered in terms of different nutritional elements, it is aimed to improve the nutritional properties of the widely produced mung bean.

Method: Optimization and characterization of the germination conditions of mung bean, a grain with high protein and fiber content, was carried out within the framework of functional and technological features. Mung beans, which were germinated under optimum conditions, were freeze-dried and turned into flour. The possibilities of using the obtained sprouted mung bean flour in beef meatballs were investigated. Meatballs, a meat product widely consumed in the world, may have negative effects on health as a result of frequent consumption due to the high amount of animal fat. In this study, the binding properties of sprouted mung bean flour, which is known to have high fiber, bioactive component and oligosaccharide content, in the formulation of beef meatballs were examined. For this purpose, L*, a* and b* values and cooking properties of the prepared and cooked foods were determined.

Results: The germination conditions of mung bean flour were optimized and the effects of its addition to the meatball composition on product quality were examined. The use of mung beans did not negatively affect the cooking efficiency and showed color characteristics close to the control group.

Conclusion: It can be said that mung bean, which has a place in people's consumption habits, is a flour that can be used by germinating and contributing to the functionality of meat products.

Effect of Food Processes on Sulforaphane content of Broccoli

Aybike Kamiloğlu¹

¹Bayburt University

Aim: It is known that broccoli has positive effects on health. This vegetable, whose consumption began to become widespread after the 1920s, has been proven to reduce some types of cancer. Sulforaphane, considered the most powerful anti-cancer agent, is the main isothiocyanate found in broccoli. Sulforaphane is released from the hydrolysis of glucoraphane by the myrosinase enzyme. The amount of Sulforaphane in broccoli may vary depending on the interaction of Glucoraphanin and myrosinase enzyme during food processing. It is important to evaluate the food processing conditions to be applied to provide food with high biological activity. Therefore, in this review, the effects of the treatments applied on the sulforaphane content of broccoli and the prevention mechanisms will be included.

Method: The effects of the applied processes will be discussed using 93 studies in the Web of Science database examining the effects of different food processing methods on the sulforaphane content of broccoli between 2000 and 2024.

Results: Although broccoli is a vegetable that can be consumed without any processing, it can undergo different processes according to consumer preferences. Broccoli is generally consumed by applying heat treatments such as boiling, steaming and microwaving. Disintegrations at the cellular level that may occur in the broccoli structure during the applied processes may cause the substances that are effective in the formation of sulforaphane to move away from the structure, and the level of sulforaphane may be affected as the increase in temperature during the processes will affect the activities of the enzymes.

Conclusion: The applied processes show that the preservation of enzyme activity is very effective on the presence of sulforaphane. While processes such as freezing and heating reduce sulforaphane formation because they limit enzyme activity, the presence of processes that increase enzyme activity supports sulforaphane formation. This review provides important information about the processes that can be preferred when consuming broccoli, which has high bioactivity against cancer.

Upcycling blueberry pomace into functional polysaccharides and their corresponding oligomers.

Ph.d. Student Vanessa Maakaroun, Salwa Karboune

¹McGill University

Aim:

Blueberries are increasingly utilized in the food industry, particularly valued for their juice and syrup applications. Around 20% of the fruit ends up as pomace after processing. Therefore, there is a critical need for developing effective approaches for producing value-added ingredients from blueberry pomace such as dietary fibers and bioactive oligosaccharides/oligomers. The pomace is primarily composed of cell wall material, which has not been extensively explored. Our study aims at investigating different extraction methods (water, alkaline, enzymatic) assisted by ball-milling or ultrasound and examining their efficiencies in terms of recovery yield, monosaccharide profile and molecular weight distribution of isolated carbohydrate extracts.

Method:

The hot water extraction was carried out at 50°C, while the alkaline extraction was achieved using 0.5, 1.5 and 2 M of NaOH. The enzymatic extraction with four different enzymes was evaluated. The assistance of these extraction by ball milling (30 Hz for 30 min) and ultrasound (40 kHz/6s on/12s off for 30 min) was carried out. The monosaccharide profile was analyzed using high-performance anion exchange chromatography, and the distribution of molecular weights was determined using high-performance liquid chromatography.

Results:

The water extraction method showed the highest recovery yield of carbohydrates (58%, w/w), followed by the enzymatic extraction (43-45%) then alkaline ones (30-41%). Assistance with mechano-milling and ultrasound enhanced the efficiency of alkaline extraction to reach a recovery yield of 68% at 1.5M NaOH and 74% at 2M, respectively. A moderate increase in the yield from 58% to 63% was observed up the assistance of the water extraction, while a decrease in the efficiency was observed for the enzymatic extraction. Several types of treatments (water, enzyme and alkaline) have displayed an increased neutral sugar content (65-90% arabinose/galactose/xylose/mannose/glucose) compared to untreated cell wall material. In terms of molecular weight, the alkaline fraction exclusively produced 10-100 kDa polysaccharides, while enzymatic and water extractions predominantly yielded 0.5-3 and 3-10 kDa oligomers.

Conclusion:

This study contributes to the efficient extraction of functional polysaccharides and oligomers from blueberry pomace. It aims to pave the way for developing well-defined carbohydrates, establishing it as a potential functional food ingredient and exploiting the untapped value of blueberry pomace.

Gut microbiome community architecture associated with fermented food consumption

Laura Judith Marcos-Zambrano¹, Blanca Lacruz-Pleguezuelos^{1,2}, Adrian Martin-Segura^{1,3}, Alberto Diaz-Ruiz³, Enrique Carrillo de Santa Pau¹

¹ Computational Biology Group, Precision Nutrition and Cancer Research Program, IMDEA Food Institute, ²Molecular Biosciences Program, UAM Doctoral School, Autonomous University of Madrid, ³Laboratory of Cellular and Molecular Gerontology, Precision Nutrition and Aging Program, IMDEA Food Institute

Aim: Fermented foods (FF) are foods or beverages produced through controlled microbial growth and the conversion of food components through enzymatic action. Although they've been part of the human diet for almost 10,000 years, their effect on the gut microbiome remains elusive. Our objective was to study the frequency of consumption of different FF in a small Spanish cohort and analyze the gut microbiome structure associated with FF consumption.

Method: We have gathered data from 70 healthy volunteers who participated in the PictureYourMicrobes study (PMID: 37530428). The data includes information on their gut microbiome (16S rRNA gene sequencing), food frequency questionnaires (including consumption of kefir, miso, kimchi, sauerkraut and kombucha), and their anthropometric measurements. Out of the 70 participants, 28 reported consuming at least one type of FF on a monthly basis. Kefir was the most popular FF among the participants (n=21), followed by kombucha (n=9) and miso (n=9). We compared the gut microbiome community structure through co-occurrence networks analysis of those who did not consume FF, those who consumed FF once a month (low consumption), and those who consumed two or more FF types on a monthly basis (average consumption).

Results: Despite alpha and beta diversity remaining similar, we did observe differences regarding the structure of the microbial community. The microbial network associated with average FF consumption appeared with a higher number of components and higher modularity (33;0.79) than those from low-FF (6;0.44) and no FF consumption (5;0.41). Moreover, keystone taxa differ between communities, appearing genera associated with SCFA production *Coprococcus*, *Prevotellaceae* UGC003, and *Faecalitalea* in average FF consumption, the beneficial *Bifidobacterium* spp. in low FF consumption network whereas the proteobacteria *Aggregatibacter* spp. in non FF consumption.

Conclusion: The inclusion of FF in the diet leads to different microbial community structures promoting the presence of beneficial microbes (SCFA producers). However, to determine the effect of FF on microbial communities and their dynamics, further research aimed at the consumption of different FF is necessary, this information is essential to reveal the role of FF in reshaping the gut microbial community and their future inclusion in dietary guidelines and recommendations.

Quantitative determination of amylase inhibitor content in beers

Silvia Matias Ibañez¹, Jon Esparta Larrakoetxea¹, Marian Bustamante Gallego^{2,3}, Jone Guenetxea Gorostiza^{4,5}, Leire Cantero Ruiz de Eguino¹, Idoia Larretxi Lamelas^{2,3}, Edurne Simón Magro^{1,2,3}, Jonatan Miranda Gómez^{1,2,3}

¹GLUTEN3S Research Group, Department of Pharmacy and Food Sciences, Faculty of Pharmacy, University of the Basque Country (UPV/EHU), ²Gluten Analysis Laboratory of UPV/EHU, Department of Pharmacy and Food Sciences, Faculty of Pharmacy, University of the Basque Country (UPV/EHU), ³Bioaraba Health Research Institute, Nutrition and Food Safety group, Araba University Hospital, ⁴Department of Preventive Medicine and Public Health, Faculty of Pharmacy, University of the Basque Country (UPV/EHU), ⁵Biogipuzkoa Health Research Institute, Mental Health Group and Psychiatric Care

Aim:

Amylase-trypsin inhibitors (ATIs) could act as activators of gastrointestinal effects and trigger symptoms in the population with functional bowel disorders, including, non-coeliac wheat sensitivity, irritable bowel syndrome, Chron's disease, or coeliac disease. ATIs constitute a protein group present in the seeds of diverse cereals, such as wheat, barley, rye, maize, millet, and rice. Beer, commonly derived from barley malt, stands as a potential source of ATIs. This beverage is consumed occasionally or regularly by approximately 81% of the population aged between 18 and 65 years. In this context, the aim of this work is to quantify the content of ATIs in beers available in the Spanish market.

Method:

Due to the fact that ATIs are alpha-amylase inhibitors, the determination of the presence of these molecules in beer was performed by an indirect method: the inhibition of alpha-amylase activity. The assay protocol was adapted from Sagu *et al.*, 2020. A maltose calibration line was used to determine the residual activity and the amount of inhibitor was quantified based on an ATI standard from *Triticum aestivum*. A 1 mg/mL of acarbose solution was used as positive control of inhibition. Twenty beers of the most popular brands in the Spanish market were analysed and the determinations were made in duplicate. Samples were diluted 1:5 to be analysed.

Results:

The results showed that the percentage of inhibition of the beers ranged from 31.9% to 71.2%, which means that ATI content ranges between 15.1 and 71.9 mg ATI equivalents/100 mL of beer. The data did not follow a normal distribution, with a median of 24.5 mg ATI equivalents/100 mL of beer and an interquartile range of 10.9 mg ATI equivalents/100 mL.

Conclusion:

In conclusion, this assay proves to be effective for the quantification of ATIs within this particular matrix. Our findings indicate varying levels of ATIs in beers, suggesting the importance of individualized analysis for each beer type. Such disparities should be considered when formulating recommendations regarding beer consumption among individuals with functional bowel disorders.

Innovative natural functional ingredients from olive oil and artichoke by-products in beef burgers

Pablo Ayuso¹, Jhazmin Qhizpe¹, Maria Angeles Rosell¹, Rocio Peñalver¹, **Professor Gema Nieto**¹

¹Universidad de Murcia

Innovative natural functional ingredients from olive oil and artichoke by-products in beef burgers

Aim:

The main objective of this study was to evaluate the antioxidant effects of artichoke in meat products through its incorporation in fat-replaced beef burgers.

Method:

Beef burgers were elaborated following 4 different formulations, a control burger; a burger with 50% of the fat replaced by an emulsion using AOVE and Prosella; a fat-replaced burger with 2.5% artichoke by-product extract; and a fat-replaced burger with 2.5% enzymatically treated artichoke extract. In order to evaluate the benefits of the reformulations, the nutritional composition, color, pH, antioxidant capacity (FRAP, ABTS and DPPH), total phenolic compounds, lipid oxidation (TBARS) and sensory attributes of the burgers were evaluated.

Results:

The results showed that the reformulations did not negatively affect nutritional parameters and sensory attributes. On the other hand, both the replacement with AOVE and the addition of artichoke significantly increased the antioxidant capacity values in the three techniques evaluated (FRAP, ABTS and DPPH). The burgers with enzymatically treated artichoke extract also presented the highest values of total phenolic compounds (445.42 mg GAE/100g). This increase in antioxidant activity resulted in a reduction of oxidation after refrigerated storage, measured through color changes and lipid oxidation (TBARS).

Conclusion:

As a conclusion, both the replacement of animal fat with AOVE and the incorporation of artichoke by-product proved to be a natural alternative to reduce oxidation of beef burgers. Potentially promoting positive effects for the consumer and not negatively affecting sensory qualities of the final product.

Effect of pre-treatments on Dietary fibre Components and Phenolic Compounds of Pearl Millet and Cowpea

Mrs Sunera Nurmomade^{1,2}, Santanu Basu¹, Irene De Carvalho², Maria Eduardo², Roger Andersson¹

¹Swedish University of Agricultural Sciences, ²Eduardo Mondlane University

Pearl millet and cowpea are consumed as a staple food in Mozambique. Dietary fibre components and phenolic compounds are two plant food constituents that are associated with many health benefits for humans.

Aim: Determine the effect of seed pre-treatment (soaking, germination and natural fermentation) on dietary fibre components and phenolic compounds in pearl millet and cowpea

Methods: Dietary fibre and its components were quantified by the Uppsala method modified for separate measurements of extractable and unextractable dietary fibre. Phenolic compounds were analysed by High-Performance Liquid Chromatography (HPLC) in reversed-phase (C18) with a diode array detector.

Results: The results indicated that soaking, germination and natural fermentation resulted in higher extractability of dietary fibre fractions in both pearl millet and cowpea compared to the washed samples. The higher extractability was seen in germinated pearl millet (6%) compared to washed pearl millet (4%). Moreover, the same pattern was seen in cowpea, germination increased the extractability in comparison to washed cowpea from 4% to 5%. Cowpea samples had a significant ($p < 0.05$) higher total dietary fibre content varied from 8.0 to 13.5 % of DM than pearl millet, which varied from 6.8 to 7.9 % of DM. For phenolic compounds, the effect on phenolic acids differed a lot between treatments. Germination and natural fermentation of both pearl millet and cowpea resulted in the formation of new compounds, this could have been due to the *de novo* synthesis of phenolic compounds that may occur on some phenolics during pre-treatments.

Conclusion: The germination and fermentation pre-treatments had a significant impact on dietary fibre components and phenolic compounds. These changes could be associated in improving the functionality of these food products and also promoting health benefits.

Matrix and pH effect on Vitamin D3 bioaccessibility and bioavailability

Evangelia Pasidi¹, Patroklos Vareltzis¹

¹Aristotle University Of Thessaloniki

Aim:

Vitamin D₃ deficiency is a global phenomenon. Supplementation and food fortification can be used to cope with this. Understanding the effect of the matrix as well as the physicochemical interactions during digestion in vitamin D₃ bioaccessibility and bioavailability is crucial in order to select the best approach of supplementation and food fortification.

Method:

Vitamin D₃ content was determined in eight different food matrices and supplements. These matrices were further tested using the INFOGEST protocol for in vitro digestion and the vitamin content was determined in each step. Further investigation of the effect of gastric pH was conducted to the supplement with the greatest bioaccessibility index to simulate digestion of different food matrices as well as fasting conditions. Emulsions with different physicochemical characteristics (different oils, viscosity, proteins, vitamin concentration and micelle size) were also subjected to in vitro digestion. The samples after emulsion intestinal digestion step were placed in Caco-2 cells to determine vitamin D₃ absorption. High-Performance Liquid Chromatography was used to determine vitamin D₃ content in the initial samples, after each digestion step and in the basolateral samples of cell cultures.

Results:

Foods exhibited higher bioaccessibility of vitamin D₃ compared to supplements. Additionally, vitamin D₃ retention percentage post-gastric digestion was notably higher for foods. A positive correlation was observed between increased pH levels and vitamin D₃ content during gastric digestion. Conversely, lower gastric pH levels resulted in heightened vitamin D₃ content during the intestinal stage. Furthermore, diverse physicochemical characteristics of emulsions influenced both the bioaccessibility and subsequent bioavailability of vitamin D₃.

Conclusion:

The composition of the matrix and the conditions of digestion play pivotal roles in determining the bioaccessibility and bioavailability of vitamin D₃. Our study's findings offer valuable insights into optimizing vitamin D₃ absorption through supplementation and food fortification, emphasizing the importance of considering not only food composition, but also potential interactions within the gastrointestinal tract.

Metabolic Impact of Future Food Processing (Meta-Pro)

Phd Maryam Rakhshandehroo¹

¹Danone Nutricia Research

Aim:

The project aims to define mild processing methods for plant-based ingredients/foods to preserve technical and nutritional properties, meet sensory requirements, and have a positive impact on health whilst simultaneously minimizing the environmental footprint and extensive refinements of ingredients.

Introduction:

Interest in plant-based foods is surging due to environmental awareness, health considerations, and the popularity of planetary diet. However, extracting plant-based ingredients often involves extensive processing, which can limit the sustainability potential. While food processing is often criticized, it can also have positive consequences. Mild approaches, like dry fractionation, reduce energy and water usage while enhancing ingredient functionality and digestion. Yet we still lack comprehensive knowledge about the effects of mild processing on plant-based ingredients' structure, functionality, and related health benefits.

Method:

We aim to understand how different levels of food processing (conventional vs. mild) impact sensory and metabolic responses to plant-based proteins and carbohydrates. Our investigation includes detailed structural analysis of the food matrix, macronutrient digestion using established models, human feeding trials, assessment of nutrient bioavailability, and comparison of sensory experiences.

Intended results:

- (i) Comparison of the human metabolic and sensory responses to plant-based foods produced by conventional vs. mild food processing for equivalent nutrient loads, including comparison of plant-based food products prepared from refined vs. less refined ingredients (i.e. proteins and carbohydrates).
- (ii) Comparison of the impact of conventional and minimal processing on nutrient digestion and bioavailability using in-vitro models and in-vivo human trials.
- (iii) An understanding of the relationship between metabolic responses and nutrient bioavailability across different degrees of food processing and food matrix structures and integrity.
- (iv) Quantification of the impact of food processing on markers of metabolic health and generation of new data to inform food producers on the best approaches to optimize sustainability, metabolic health, and sensory impact of their processes.

Conclusion:

The knowledge generated will be used to develop guidelines for healthier and more sustainable approaches to plant-based food processing.

Studying the digestive fate of amyloid-like fibrils in food systems

Mrs. Gil Refael¹, Dr. Yizhaq Engelberg¹, Dr. Alon Romano¹, Ms. Gabriela Amiram¹, Dr. Eilon Barnea¹, Dr. Carmit Shani Levi¹, Dr. Sondra Turjeman², Prof. Meytal Landau¹, Prof. Omry Koren², Prof. Uri Lesmes¹

¹Technion - Israel Institute of Technology, ²Azrieli Faculty of Medicine, Bar-Ilan University

Aim: Protein amyloids have been implicated in various pathogeneses, such as Alzheimer's disease, raising concerns over their formation in foods and subsequent effects on consumer health. This work delves on the possible formation of edible proteinaceous nano-architectures in food systems and their uncharted digestive fate.

Method: Firstly, evidence presented will exemplify process-induced formation of amyloid-like architectures from alpha-lactalbumin, β -lactoglobulin or ovalbumin through experimental findings of ThT analysis and TEM imaging (Refael et al., 2024; Romano et al., 2023). These stable assemblies can successfully be used as delivery vehicles for controlled release of lipophilic bioactives, such as capsaicin (Romano et al., 2023). Moreover, coupling of in vitro human digestions with LC-MS/MS proteomic analyses shows such supramolecular assemblies exhibit attenuated susceptibility to gastric and intestinal proteolysis and may persist into the colon. Therefore, human fecal fermentations of fibrils or their progenitor proteins was conducted in anaerobic bioreactor followed by 16S rRNA gene sequencing and bioinformatic analysis using QIIME2 for proportional gut community analysis.

Results: These show fibrilization preserves colonic microbial diversity, low Firmicutes/Bacteroidetes ratio and protects butyrate producing genera, namely Roseburia and Clostridium, similarly to prebiotic fructooligosaccharides. In addition, PICRUSt2 *in silico* analyses providing metabolic pathway predictions support that fibrils divert microbiota metabolic trajectories towards those observed in fermentation of prebiotics.

Conclusion: Thus, this work will provide comprehensive evidence and insights into protein amyloids in foods and their relevance to food safety and functionality. Moreover, this work will raise the question if amyloids may exhibit differential digestive fates in different consumers along the healthy life span.

Liposomes as a new delivery system for phytosterols

Professor Magdalena Rudzińska¹, Dr Anna Grygier², dr hab. Maciej Jarzębski³, dr hab. Wojciech Smółka⁴, dr hab. Katarzyna Cieślik-Boczula⁵

¹Poznań University Of Life Sciences, ²Poznań University Of Life Sciences, ³Poznań University Of Life Sciences, ⁴Poznań University of Technology, ⁵University of Wrocław

Aim: Liposomes are one type of nanoparticle that have found widespread use as deliverers for targeted drugs and as a source of functional compounds in food. They have improved therapies for a range of biomedical applications by stabilizing therapeutic compounds, overcoming obstacles to cellular and tissue uptake, and improving the biodistribution of compounds to target sites in vivo. The last two decades have seen a strong interest in liposomes in food technology and dietetics. Bioactive ingredients such as phytosterols and their esters have been found for a number of commercial and clinical applications. These compounds are susceptible to oxidation and degradation during food processing and heating. Encapsulating them within lipid-based nanocarriers makes their application in food products possible. The main goal of these research was to develop a method to obtain liposomes which encapsulate phytosterols to assess their physicochemical properties related to quality and thermo-oxidative stability. **Methods:** The standard of stigmaterol was encapsulated in liposomes. The structure of both lipid membranes of liposomes and encapsulated molecules and interactions between lipids and encapsulated components were qualitatively and quantitatively studied using Fourier transform infrared (FTIR). The analysis of thermodynamic properties of lipid membranes in the obtained liposomes were done by estimation of enthalpy, partial heat capacity, cooperativity of the phase transition associated with the differential scanning microcalorimetry (DSC) peak width for exo- and endothermic peak transitions of lipid membranes measured in different scan rates and incubation time. The thermooxidative stability of stigmaterol encapsulated in liposomes was determined after heating at 60 and 180°C. The products formed during degradation of liposomes and stigmaterol were determined using HPLC-SEC/ELSD and GC-FID methods. Stigmaterol oxidation products were identified by GC x GC/MS ToF method. **Results:** In FTIR spectra dominate the signals from DPPC phospholipid. Comparing the spectra of the initial liposomes (with and without stigmaterol), no significant differences were found, especially outside the dactyloscopy region of the spectrum. The signals specific to stigmaterol are weak or not visible. This is largely due to the fact that these additives are relatively small, and their signals largely coincide with those characteristic of DPPC. However, the effect of temperature was observed. The changes were visible only after heating at 180°C, but not at 60°C. This may suggest thermal degradation of liposome components, including oxidation, as evidenced by stronger signals from C-O bonds around 1100 cm⁻¹. These observations were confirmed by chemical analysis. The degradation of stigmaterol encapsulated in liposomes heated at 60 and 180°C was 8 and 89%, respectively. When free stigmaterol was heated at the same temperatures, its degradation was similar and amounted 7 and 80%. Degradation of phytosterols in rapeseed oils after thermo-oxidation at 60 and 180°C was 10 and 40%. In all heated liposomes stigmaterol oxidation products were determined. **Conclusion:** - The procedure of stigmaterol encapsulation in liposomes has been developed; - The yield of encapsulated liposomes was about 20%; - The encapsulation of stigmaterol in liposomes did not protect it from thermo-oxidative degradation at 180°C; - It is necessary to check the effect of the protective layer of liposomes on the stability of stigmaterol; - The stability of stigmaterol encapsulated in liposomes in digestive system ought to be analysed; - They can be very important for human health especially for people with cardiovascular diseases (CVD)

Indices that assess nutritive value and environmental impact of meals and diets; a systematic review.

Miss Eva-leanne Thomas¹, Dr Joe Livingstone¹, Dr Anne Nugent^{1,3}, Professor Jayne V. Woodside^{2,3}, Professor Paul Brereton^{1,3}

¹School of Biological Sciences, Queen's University Belfast, ²School of Medicine, Dentistry and Biomedical Sciences, Queen's University Belfast, ³The Institute for Global Food Security, Queen's University Belfast

Aim:

The aim of this systematic review is to determine what food indices exist for classifying or ranking meals or diets in terms of both human and planetary health. It will outline the methodologies and key characteristics of such indices and identify their intended public health purpose.

Method:

This systematic review was carried out following the PRISMA 2020 statement: an updated guideline for reporting systematic reviews. Six bibliographic databases were searched using keywords and medical subject headings (MeSH) related to 'nutrition', 'environment', 'health', 'index', 'meal' and 'diet'. Records of retrieved studies were screened independently by two reviewers in Rryan. Data extraction included but was not limited to index name and dimensions, target population, index aim, public health purpose, name of nutritional and environmental scores, list of nutrient values, list of environmental impact indicators, cut-off values for both nutritional and environmental scores independently, system boundaries, and cut-off values for healthy and environmentally friendly meals or diets.

This work assessed nutritive value and environmental impact food indices in the context of meals and diets for the first time.

Results:

There are an increasing number of indices assessing both nutritive value and environmental impact of meals and diets within the peer-reviewed literature. 5,946 papers were retrieved from the search strategy after duplicates were removed. The index characteristics, methodology and purpose will be presented together with an assessment of its performance.

This systematic review can be used to inform researchers, business, and policy actors on potential future approaches for labelling foods, meals or diets in a way that reflects both human and planetary health, e.g. adaptation of public health policies or initiatives, such as highlighting the need for meal reformulation or marketing restrictions.

Conclusion:

This systematic review highlights the variety of food labelling indices that exist assessing nutritive value and environmental impact of meals or diets. This can be used to assist public health actors in the future development of an index to support healthy and sustainable meal and diet choices.

Prebiotic effect of Moringa Oleifera on Gut Microbiota Composition

Ph.d Lidia Tomás-Cobos¹, Elisa Gallego Vendrell¹, Elena Díez-Sánchez¹, Blanca Viadel Crespo¹, Nataly Peña-Gómez², Juan David Escobar-García²

¹AINIA, ²Q'omer BioActive Ingredients S.L.

Aim: The alteration in the gut microbial community has been regarded as one of the main factors related to metabolic disorders. To date, little is known about Moringa oleifera as a nutritional intervention to modulate the dysbiosis of intestinal microbiota. Therefore, this study aimed to explore the prebiotic effect of Moringa oleifera leaf extract on intestinal microbiota, using a miniaturized in vitro model of colonic fermentation.

Method: To study the prebiotic effect of the Moringa oleifera leaf extract, an in vitro miniaturized system of colonic fermentation was used. The system reproduces the gastrointestinal and colonic metabolism of Moringa oleifera leaf extract. The in vitro miniaturized system of colonic fermentation was inoculated with faecal microbiota from adults. Intestinal microbiota modulation was studied by analyzing the main microbial groups using selective media and direct plate culture methods. To study the effect on the microbial colonic composition, aliquots of colonic digest have been taken to analyze the entire composition of the colon microbiota by means of 16S metagenomic analysis. Moreover, the metabolic activity of the intestinal microbiota was evaluated through the analysis of short-chain fatty acids (SCFAs) production in the colonic digested samples by Gas Chromatography with FID detector.

Results: The results obtained in the present study show that the Moringa oleifera leaf extract produces a significant increase in viable beneficial bacteria of the genus Lactobacillus in the colonic microbiota after 3 days of colonic fermentation. In addition, the results obtained for the colonic microbiota with the 16S metagenomic analysis show that Moringa oleifera leaf extract produces an increase in healthy bacteria of the genera Lactobacillus and Bifidobacterium, and of the bacteria Bacteroides and Escherichia/Shigella not associated with pathogenic effect. Finally, was observed a positive influence on the metabolic activity of the colonic microbiota, due to an increase in total SCFAs, mainly due to acetic acid and propionic acid.

Conclusion: Moringa, rich in fibre and protein, promotes the growth of beneficial bacteria of the genera Bifidobacterium and Lactobacillus in the human colonic microbiota, and produces an increase in the concentration of total SCFAs (healthy metabolites). With this, the beneficial effect of Moringa (prebiotic effect) on the colonic microbiota and its metabolic activity has been scientifically evidenced.

Strategic choices in preserving the health potential of vegetables when prepared in industrial kitchens

Flore Vancoillie¹, An Callens², Kaat Vanhegen², Sarah H.E. Verkempinck¹, Tara Grauwet¹

¹KULeuven, ²VIVES

Aim:

Many people rely on meals prepared in industrial kitchens (e.g. schools, elderly homes, hospitals, home delivery, etc.). Their ultimate goal is to deliver microbial-safe, convenient and warm food on a large scale. However, these meals are generally low in vegetable content and their nutritional quality is often neglected. Therefore, health-related compounds and their corresponding (bio)chemical conversions were evaluated in leek (i.e. vitamin C, S-alk(en)yl-L-cysteine sulfoxides (ACSOs)) and Brussels sprouts (i.e. vitamin C, glucosinolates) during different industrial kitchen approaches existing today.

Method:

Based on surveys in Belgian hospitals and large-scale catering services, two commonly used industrial approaches were mapped out: (i) steaming followed by a holding time for maximally 3 h at 80 °C until serving ("single-heat-preparation"), or (ii) steaming followed by cold overnight storage and reheating via microwaves (portioned) or convection (in bulk) followed by a holding time for maximally 1 h at 70 °C until serving ("double-heat-preparation"). The impact of each step of these approaches was evaluated on (i) freshly diced leeks and (ii) industrially frozen Brussels sprouts, to include differences in pre-preparation processing steps. In addition, both their (i) intact and (ii) pureed forms were evaluated, to include differences in structure, meeting the demand for the vegetable foods lower need for in-mouth processing (e.g. to account for swallowing difficulties).

Results:

In freshly diced leeks, pureeing caused almost complete enzymatic conversions of ACSOs into health-beneficial organosulfur compounds and degradation of vitamin C. No major glucosinolates and vitamin C conversions could be established in Brussels sprouts, as industrially blanching before freezing already eliminated corresponding enzymes. In both matrices and in both approaches, vitamin C concentrations were strongly impacted by each heating step resulting in a large or even complete reduction at the final holding time. However, impact was smaller for Brussels sprouts due to previous inactivation of ascorbic acid oxidase. ACSOs and glucosinolates remained majorly stable through both preparation approaches.

Conclusion:

Overall, the highest health-related quality was obtained for the single-heat-preparation method. Long holding times were detrimental to vitamin C concentrations. It was concluded that matrix, structure and pre-processing had an impact on health-related compound stability through preparation.

How anthocyanins affect the gut microbiome and their impact on inflammatory process: a bibliometric analysis

Juliana Dara Rabêlo Silva¹, Raquel Bedani¹, **Douglas Xavier dos Santos**^{1,2}, Henrique Silvano Arruda¹, Amanda Cristina Andrade¹, Paulo César Martins Alves³, Glaucia Maria Pastore¹, Anderson S. Sant'Ana¹, Adriane Elisabete Costa Antunes⁴, Katia Sivieri⁵, Mário Roberto Maróstica¹

¹Department of Food Science and Nutrition, Faculty of Food Engineering, University of Campinas,

²Fraunhofer Institute for Process Engineering and Packaging (IVV), ³Center for Investigation in Pediatrics (CIPED), School of Medical Sciences, University of Campinas, ⁴School of Applied Sciences (FCA), University of Campinas, ⁵Department of Food and Nutrition, School of Pharmaceutical Sciences, São Paulo State University (UNESP)

Aim: The objective of this work is to provide perspectives on the prebiotic potential of anthocyanins on the modulation of the gut microbiome and its impact on inflammatory diseases.

Method: One hundred and forty studies published in the Clarivate Analytics' Web of Science database from 2015 to 2024 were evaluated and performed to a bibliometric analysis employing the VOSviewer v.1.6.19 (<https://www.vosviewer.com/>) for scientific mapping and network analysis.

Results: A bibliometric mapping plotted from 37 terms present in the title/abstract originated four groups (red, green, blue, and yellow) through software (Figure 1). It is interesting to note that the red cluster highlights 14 terms related to studies *in vivo* that evaluated the impact of anthocyanins consumption on oxidative stress, insulin resistance, and obesity. The green cluster showed 11 items focused on the microbiota modulation and synthesis of short-chain fatty acids in studies with animal models. The blue cluster possesses 9 terms associated with inflammatory bowel disease and biomarkers of inflammation. Finally, the yellow cluster concentrated 3 terms relative to the use of cyanidin 3-glucoside in the studies. Moreover, the top 10 terms most frequent in title/abstract in the function of the number of occurrences are "microbiota" (n = 81), "mechanism" (n = 33), "obesity" (n = 31), "abundance" (n = 31), "high fat diet" (n = 29), "consumption" (n = 27), "day" (n = 26), "rat" (n = 25), "cyanidin" (n = 24), and "oxidative stress" (n = 24). The terms "microbiota", "mechanism", and "obesity" represent 43.8% of these occurrences. Our results suggest that the administration of anthocyanins influences the modulation of the gut microbiome and is associated with a reduction of biomarkers of inflammation. Since these bioactive compounds demonstrated the relief of symptoms of some inflammatory diseases.

Conclusion: Therefore, bibliometric analysis demonstrated that anthocyanins possess prebiotic potential in terms of modulating the gut microbiome, as well as contributing to the anti-inflammatory effect through the synthesis of short-chain fatty acids by the beneficial microbial community. Future perspectives point to the modulation of the gut microbiome by anthocyanins supplementation as a promising adjuvant in improving symptoms associated with inflammatory diseases.

How probiotics, prebiotics, synbiotic and postbiotics might collaborate to feminine health: a bibliometric analysis

Douglas Xavier-Santos¹, Raquel Bedani¹, Isabel de Almeida Vieira², Marina Padilha³, Clara Mariana Gonçalves Lima¹, Juliana Dara Rabêlo Silva¹, Beatriz Manfrinato Ferreira¹, Paulo César Giraldo⁴, Jorge Pamplona Pagnossa⁵, Katia Sivieri⁶, Adriane Elisabete Costa Antunes², Anderson S. Sant'Ana¹

¹Department of Food Science and Nutrition, Faculty of Food Engineering, University of Campinas,

²School of Applied Sciences (FCA), University of Campinas, ³Department of Social and Applied

Nutrition, Federal University of Rio de Janeiro, ⁴Department of Obstetrics and Gynecology, School of Medical Sciences, University of Campinas, ⁵Department of Biological Sciences, Pontifical Catholic

University, ⁶Department of Food and Nutrition, School of Pharmaceutical Sciences, São Paulo State University (UNESP)

Aim: The objective of this work was propitiated insights and conception about the influence of probiotics, prebiotics, synbiotic, and postbiotics as adjuvants for prevention/treatment on the main pathologies that can affect women's health.

Method: Seventy-one studies published in the Clarivate Analytics' Web of Science database from 1999 to 2024 were evaluated and performed to a bibliometric analysis employing the VOSviewer v.1.6.19 (<https://www.vosviewer.com/>) for scientific mapping and network analysis.

Results: A bibliometric mapping plotted from 39 keywords considering with a minimum number of two occurrences of which six of them had an average of more than 40 citations ("aerobic vaginitis", "bacterial vaginosis", "chlamydia trachomatis", "lipid profiles", "polycystic ovary syndrome", and "synbiotic"). The top 10 author keywords with the highest number of occurrences (larger spheres) are "probiotics" (n = 20), "lactobacillus" (n = 13), "vaginal microbiota" (n = 12), "probiotic" (n = 12), "lactobacilli" (n = 12), "bacterial vaginosis" (n = 10), "polycystic ovary syndrome" (n = 6), "biofilm" (n = 5), "candida" (n = 4), and "dysbiosis" (n = 4). Moreover, The top 10 author keywords with the highest number of occurrences (larger spheres) are "probiotics" (n = 20), "lactobacillus" (n = 13), "vaginal microbiota" (n = 12), "probiotic" (n = 12), "lactobacilli" (n = 12), "bacterial vaginosis" (n = 10), "polycystic ovary syndrome" (n = 6), "biofilm" (n = 5), "candida" (n = 4), and "dysbiosis" (n = 4). Our results suggest that the administration of biotic agents as adjuvants are relevant for the prevention and/or treatment of the main pathologies that affect female health, since they contribute to a healthy vaginal microbiota through anti-inflammatory and antimicrobial activities.

Conclusion: Therefore, these clinical studies have demonstrated the effectiveness of intervention using probiotics to the detriment of other biotic agents in feminine health, being bacterial vaginosis, polycystic ovary syndrome, and vulvovaginal candidiasis, the main pathologies evaluated. Future perspectives point to the beneficial modulation of the vaginal microbiota by biotic agents as a promising adjuvant approach to improve women's health.

Phosphatidylcholine regioisomer composition in rat organs feed with n-3 deficiency and structured triacylglycerol diets

Msc Yuqing Zhang¹, Baoru Yang, Mikael Fabritius, Guðmundur Haraldsson, Haraldur Guðmundsson

¹University of Turku

Aim: (heading must be in bold)

(Introduction text - align left, 10 point, Times New Roman, single line spacing)

As structural and functional components of cell membranes, glycerophospholipids (GPLs) play a crucial role in a wide range of physiological processes. Positional distribution of fatty acid (FA) on PL molecular greatly influences the quality, nutritional properties, and physiological effects of fats and oils. This study aims to establish a comprehensive overview of the PC species distribution in rat organs. In addition, to reveal the bioavailability and the metabolic fate of the valuable n-3 FAs, an animal trial was conducted with rats feeding with structured TAGs containing DHA in different positions.

Method: (heading must be in bold)

(Methods text - align left, 10 point, Times New Roman, single line spacing)

This study thoroughly investigated the phosphatidylcholines (PCs) regioisomeric composition in organs (liver, brain, heart, spleen, lung, testicle, kidney, and eye) of rats fed with different structured triacylglycerols (TAGs), employing a novel tandem mass spectrometric (MS/MS) methodology coupled with a computational algorithm.

Results: (heading must be in bold)

(Result text must - align left, 10 point, Times New Roman, single line spacing)

A total of 133 different PC regioisomers were detected in the eight organs. Our findings reveal differences between each organ in some PC species and regioisomers pairs. It has been proved that in natural fats, the unsaturated FAs tend to esterify on the sn-2 position on PL molecular, but our results showed that is not always the case. Most notably when two UFAs have a double bond number difference less than 2. For example, PC 22:6/20:4 is always the dominant regioisomer in the pair instead of PC 20:4/22:6 in all organs researched.

Conclusion: (heading must be in bold)

(Conclusion text must - align left, 10 point, Times New Roman, single line spacing)

Our study demonstrated that four-week n-3 deficient feeding changed the composition and the relative abundance of PC species and regioisomers. Supplementation with docosahexaenoic acid (DHA) in regio- and stereospecifically structured TAGs not only counteracted the changes in PCs induced by n-3 deficient feeding but also resulted in higher number and relative abundance of DHA-containing species compared to n-3 adequate normal feed.

Comparison of phospholipid regioisomers in mammalian milk using liquid chromatography quadrupole time-of-flight mass spectrometry

Miss Qizhu Zhao¹, Yuqing Zhang¹, Mikael Fabritius¹, Marika Kalpio¹, Baoru Yang¹

¹Food Sciences, Department of Life Technologies, University of Turku

In mammalian milk, fat contributes to nearly half of the daily energy intake of newborns. Phospholipids (PLs) play an important role in constructing the milk fat globule membrane. As both bioactive compounds, PLs not only contribute to fat absorption, but also provide the building blocks for cell membranes and play a major role in physiological processes such as cognitive function and the regulation of inflammation. PL regioisomers, having the same fatty acids (FAs) but differing in stereospecific positions of the FAs in the molecule, may have different physicochemical properties and metabolic fates *in vivo*. However, positional isomers of PLs in mammalian milk remain largely unexplored. Previous research has mainly reported the phospholipid classes of cow and human milk, but the FA distribution and structural characteristics of PLs in other mammalian species are still unknown.

In this study, the regioisomeric composition of the phosphatidylcholine (PC) was analyzed in eight mammalian species including human, ruminants: cow, camel, goat, sheep, non-ruminants: dog, horse and pig. The recently developed method is based on reversed-phase liquid chromatography quadrupole time-of-flight tandem mass spectrometry (RPLC-QTOF-MS/MS) hyphenated with an electrospray ionization source. An in-house calculation software enabled the accurate calculation of regioisomer ratios of PCs. Nearly 30 kinds of PC acyl carbon number : double bond (ACN:DB) species were detected. The species distribution and regioisomer composition varied among different milks. PC 36:2 was the most abundant species in human, compared with PC 34:1 in cow and PC 32:0 in horse milk. Further, for example, species PC 34:1, which accounted for almost over 10 mol% in milks, mainly dominated by PC 18:1/16:0 in human, while PC 16:0/18:1 contributed more in cow, and interestingly, these two PCs distributed evenly in horse milk. Additionally, some PC regioisomer pairs were unique, such as PC 14:0_18:3 and PC 16:1_18:3 in horse milk.

This study provides a reference to find out ideal sources of PLs. Knowledge on PL regioisomer composition in mammalian milk and its contrast to human milk composition would contribute to a better understanding of their differences and nutritional values, ultimately generating ideas for future research on human milk substitutes.

The intake of Sambucus nigra extract can impact, in vitro, the human gut ecosystem

Mr. Francis Aheto¹, Dr. Andrea Polo¹, Dr. Olga Nikoloudaki¹, Miss Lena Granehäll¹, Dr. Stephan Plattner², Prof. Raffaella Di Cagno¹, Prof. Marco Gobbetti¹

¹Free University of Bozen-Bolzano, ²Iprona AG SpA

Aim:

In response to the growing interest in the health benefits of plant-based bioactive compounds, this study aimed to investigate the *in vitro* effects of an elderberry extract containing polyphenols and fiber on the human gut microbial ecosystem.

Method:

The study employed the simulator of the human intestinal microbial ecosystem (SHIME®). The setup consisted of two identical SHIME® units running in parallel, each unit including 3 bioreactors simulating the stomach/small intestine, proximal colon (PC), and distal colon (DC), respectively. Each unit was inoculated with feces from a different representative healthy donor adhering to the Mediterranean diet. The experiment comprised: (i) a stabilization of colon ecosystems; (ii) a two-week control period (baseline); (iii) a two-week treatment period in which 600 mg/day of elderberry extract was added to the diet; and (iv) a one-week washout period to assess the persistence of effects. Lumen samples were collected from colon bioreactors at the end of the control period, after one and two weeks of treatment period, and at the end of washout period. Samples were analysed for short-chain fatty acid (SCFA) profiles, and structure and potential functionality of bacterial communities using HPLC-UV and shotgun metagenomics.

Results:

The treatment did not impact the overall profiles of acetic and butyric acids. Differently, propionic acid significantly increased after the extract intake in both colon tracts of only one donor. While the donor and the colon tract influenced overall microbiome composition (beta diversity), the treatment significantly modulated species diversity (alpha diversity). The administration of elderberry extract increased, in at least one donor, the abundance of several bacterial species having the potential to exert beneficial activities. Moreover, several unwelcome species decreased. Potential functionality analysis demonstrated that the treatment had significantly increased genes related to amino acid metabolism, and gut barrier function in PC of donor 1, with effects persisting after the washout, while donor 2 exhibited fewer significant changes.

Conclusion:

The elderberry extract affected the gut ecosystem mainly by modulating bacterial microbiota composition and putative function in a donor-dependent manner. Further research is needed to understand the long-term effects and mechanisms of action.

Innovative modification of antioxidant capacity assays to address specific problems in food chemistry

Professor Reşat Apak^{1,2}, Burcu Bekdeşer¹, Mustafa Bener³, Saliha Esin Çelik¹, Sema Demirci Çekiç¹, Ayşem Arda¹, Ziya Can¹, Şener Sağlam¹

¹Istanbul University-Cerrahpaşa, Faculty of Engineering, Chemistry Department, Analytical Chemistry Division, ²Turkish Academy of Sciences (TUBA), Bayraktar Neighborhood, Vedat Dalokay Street No: 112, ³Istanbul University, Faculty of Science, Department of Chemistry, Analytical Chemistry Division

Aim:

Antioxidant capacity (AC) assays are usually based on electron-transfer reactions measuring the reducing capability of polyphenols, carotenoids, thiols and vitamins toward chromogenic probes like CUPRAC or ABTS reagents. However, most antioxidants behave as scavengers of reactive species (*i.e.* ROS, RNS) in addition to electron donation, and the extent of their radical quenching may be thermodynamically or kinetically unpredictable. Some antioxidant probes may be immobilized on membrane substrates or interact with nanoparticles to meet the specific needs of the interface between human health and nutrition. The aim is to modify established assays for problem solving.

Methods:

The CUPRAC reagent, Cu(II)-neocuproine, was either immobilized on a polymeric sulfate/sulfonate membrane or held on heparine-modified gold nanoparticles (AuNPs), on which antioxidants produced the orange Cu(I)-neocuproine chelate. The CUPRAC reagent responded to selected probes that have undergone the attack of hydroxyl/superoxide radicals. Thiol-disulfide mixtures could be assayed for both constituents using a single reagent (ABTS^{•+}) after incubating the mixtures at 60°C for 60 min. The AuRAC assay was based on the absorption measurement of the bluish-green oxidized product (ABTS^{•+}) formed as a result of the oxidation reaction of ABTS reagent with gold (III).

Results:

When the CUPRAC reagent was adsorbed on membranes or heparinated AuNPs, cuprous-neocuproine formed *in situ*, enabling measurement in turbid food matrices. In measuring reactive species or their oxidized products, the change in the CUPRAC absorbance of either the starting probe or its oxidized product was measured to indicate reactive species; some of this absorbance difference was recovered with antioxidants to enable AC determination. The problems of preliminary operations arising from direct borohydride reduction of disulfides to thiols were eliminated with the use of a single reagent (ABTS^{•+}) after incubation with thiol-disulfide mixtures. The AuRAC assay together with ABTS colorimetry enabled AC determination by attenuation of the color arising from ABTS^{•+} without AuNPs formation.

Conclusion:

Conventional AC measurement methods have been modified to address specific problems, of which the experimental and theoretical outputs meet the need to surpass the conventional functions of AC reagents to achieve certain goals in food and biological chemistry.

The Sus-Health index: composition, presentation and application to meals

Dr Joe Livingstone¹, Dr Vasilis Grigoriadis², Professor Paul Brereton¹, Professor George Hutchinson¹, Dr Beatrice Smyth¹, Dr Jelena Vlajic¹, Dr Anne Nugent¹, Professor Jayne Woodside¹, Miss Eva Leanne Thomas¹, Dr Francisco Areal⁴, Dr Orla Collins³, Dr Novieta Sari³, Dr Rao Fu³, Dr Ransford Teng-viel Karbo³, Professor Lynn Frewer³

¹Queen's University Belfast, ²University of Ioannina, ³Newcastle University, ⁴Northumbria University

Aim:

The aim of this work is to produce a novel single index (the Sus-Health index) that describes both the nutritive value and environmental impact of meals to encourage consumers to make improved food choices regarding human and planetary health.

Method:

The Sus-Health index was co-developed over an 18-month period with the synergy of different parts of the UK food supply chain (e.g., restaurants, governmental and local authorities, wholesalers). Designed specifically to assess meals, but applicable to individual foods and ingredients, the Sus-Health index is a product of existing EnviroScore and Nutrient Profiling Model (NPM) indices. EnviroScore was calculated using SimaPro 9 software and its databases. NPM was calculated using the Nutritics data base. The two separate indices were combined using the composite indicator methodology and weighting of 1:1. Results can be presented quantitatively or qualitatively on a scale of A (best score)-E (worst score). Presentation of the index was assessed using consumer studies. The index was applied to 8 meals from a local restaurant using their recipe and supply chain information. The meals included vegan, meat and seafood options.

Results:

Consumer studies indicated that presentation on menus as a “doughnut” was favoured with the overall score colour coded in the “core” together the two individual components in a colour coded surround. A range of A-D scores were found for the 8 meals assessed. Although the index was co-created with relevant stakeholders it was acknowledged that it is dependent on the limitations of the component parts: 1) NPM is restricted to a specific range of nutrients (e.g. does not consider specific micronutrients); 2) Calculations for assessing the Environmental Impact component invariably use highly aggregated data.

Conclusion:

This work aims to help facilitate the transition to a more sustainable food system by proposing an innovative measure; the Sus-health index to inform consumers, allowing for improved food choices. The index has been used to assess and classify the scores of different foods and meals. The index is to be assessed in future Living Lab experiments in restaurants to assess its impact on consumers in terms of making more healthy and sustainable choices.

Symbiotics in the metabolic syndrome and obesity; the example of *Yarrowia Lipolytica*

Dr PATROKLOS VARELTZIS¹, Dr Sotirios Patsios²

¹Aristotle University of Thessaloniki, ²The Center for Research and Technology Hellas

Aim: The microbiomes of obese individuals are structurally and functionally different, strongly suggesting the microbiome as a potential target for the prevention and/or treatment of obesity. Probiotics and prebiotics have emerged as effective and integrated means to regulate the microbiome in order to reverse the microbial dysfunction associated with the obesity phenotype. Symbiotic foods may be a vehicle in this effort. This presentation summarizes the latest research developments on the role of symbiotic foods in obesity and the technological challenges of their production. Finally, the recent results of our laboratory research on improving *Yarrowia Lipolytica* viability during processing for potential use as probiotics in foods are presented.

Method: *Yarrowia Lipolytica* MUCL 28849 was cultivated in a 3L bioreactor containing YPG medium. Following a 48-hour incubation period under conditions of pH 6, 30°C temperature, 1vvm sterilized air, and stirring at 800rpm, the cells were harvested via centrifugation. Subsequently, the cells were washed with PBS and suspended in sterile distilled water at a concentration of 10% w/v. Inulin and maltodextrin were assessed as potential wall materials during the spray drying process using an ADL311S spray dryer. Various parameters including temperature, drying air velocity, and feed rate were investigated during experimentation. The dried cells were then subjected to evaluation for their viability, carotenoid content, and antioxidant capacity. Additionally, a simulated digestion process based on the INFOGEST protocol was employed to assess the viability of the cells throughout different stages of digestion.

Results: The highest post-spray drying cell viability was observed in the powder encapsulated with inulin, suggesting its ability to create a protective microenvironment around the cells. Maltodextrin also provided significant protection, particularly at a concentration of 10% w/v, with notable effectiveness observed at a drying temperature of 130°C. Following simulated digestion, cells encapsulated in maltodextrin exhibited substantially higher viability compared to those encapsulated in inulin.

Conclusion: The findings indicate that spray-dried *Y. lipolytica* yeast holds promise for application in food products due to its favorable nutritional profile, excellent viability post-drying, resilience during simulated digestion, and potential probiotic activity.

Long-term memory of experts for the definition of wine aroma descriptors of resistant varieties

Tomas Roman¹, **Nicola Cappello**¹, Demetrio Dell'Oca², Andrea Natolino³, Emilio Celotti³, Roberto Larcher¹, Adelaide Gallo^{1,2}

¹Fondazione Edmund Mach - Center for Technology Transfer, ²C3A – Centro agricoltura Ambiente,

³Università degli Studi di Udine—Dipartimento di Scienze Agroalimentari, Ambientali e Animali

Aim:

This work aimed to elaborate a list of descriptors by aroma classes and subclasses of the mold-resistant hybrid varieties Sauvignier gris (SG) and Solaris (SOL) wines.

Method:

A free-word survey was carried out to pinpoint the predominant aroma descriptors of Sauvignier gris (SG) and Solaris (SOL) wines, based on the long-term memory of expert tasters of each variety. Participants comprised of twenty-five Sauvignier gris and twenty Solaris producers/experts from five Italian wine regions. Experts were posed with the following question: *"How would you describe the aroma of Sauvignier gris/Solaris?"* No additional information or prior knowledge about the wine style, production protocols, or harvest was considered.

Results:

Experts proposed 115 unique descriptors for SG and 96 for SOL. The descriptors were lemmatized and categorized into classes and subclasses according to the aroma wheel originally proposed by Nobel et al. (1984), and subsequently refined. For both varieties, over 90% of the descriptors were directly included or were synonyms comprised in the aroma wheel. The remaining were addressed in the different classes and subclasses through a triangulation process conducted by three winemakers. Six classes of aroma were identified for both varieties. The experts surveyed reported that the fruity descriptors were the most frequently recalled, constituting 49% of those of SG and 55% of SOL. Nevertheless, differences among varieties were substantial. Among the six subclasses grouping fruity descriptors, the most frequently occurring for SG was "Undefined fruity" (27%), followed by "Tropical" (21%) and "Citrus-like" (16%). In contrast, SOL was predominantly characterized by "Tropical" descriptors (42%), followed by "Citrus-like" (25%) and "Undefined fruity" (11%). "Vegetative" descriptors were more frequent in SG (20%) than in SOL (11%), and for both varieties, these were mostly associated (~50%) to the "balsamic/aromatic herbs" subclass. The percentage of "Floral" descriptors was approximately 10% for both varieties, with over 85% of them categorized under the "White flowers" subclass. The frequency of the "Aging", "Defect", and "Mineral" classes was ≤ 10%, being the latter more represented in SOL (10%) than in SG (4%).

Influence of flavouring application technique, flavour and hydrocolloid type on sensory perception of 3D-printed snacks

Kristina Radoš, **Prof Nikolina Cukelj Mustac**¹, Marija Topić, Bojana Voučko, Dubravka Novotni

¹University Of Zagreb Faculty Of Food Technology And Biotechnology

Aim: While the flavouring of conventionally produced snacks is well developed, flavouring of 3D-printed snacks remains insufficiently addressed. Thus, we investigated how different approaches of flavour addition and variation of hydrocolloids influence sensory experience of gluten-free 3D-snacks.

Method: Gluten-free batter (millet flour/sweet potato flour/rice protein) with 1.8% psyllium was flavoured with (i) dry onion powder (PSY-SPICE), (ii) onion powder flavouring (PSY-FLAVOUR) or (iii) solution of onion powder flavouring sprayed onto printed batter (PSY-SPRAYED). The aim was for the final onion spice/flavour amount to be 2% (w/w) of the powdered ingredients. Batter was extrusion-printed into a 12-layer cloud-shape, and oven-dried (120°C/30 min). In the second phase, onion flavouring was added to batter with psyllium omitted (CTRL-FLAVOUR) or replaced with 2% sodium alginate (NaA-FLAVOUR). Fourteen panellists participated in the Just About Right test (JAR), descriptive (0-10 scale) and hedonic (1-9 scale) sensory evaluations.

Results: Onion odour intensity as well as overall odour were similar for PSY-SPICE (5.1±1.7; 5.7±1.2) and PSY-FLAVOUR snacks (4.8±1.1; 5.8±0.9) but significantly lower in PSY-SPRAYED (3.2±1.7; 3.8±1.4). In addition, onion flavour intensity was lowest in PSY-SPRAYED (3.9±0.9) and highest in PSY-FLAVOUR (5.4±0.9). Flavouring technique did not affect saltiness or overall flavour perception. In the JAR test, 9 and 8 panellists found the onion flavour just right in PSY-SPICE and PSY-FLAVOUR, respectively, while 10 deemed it too little in PSY-SPRAYED. Consequently, 7 and 6 panellists preferred PSY-SPICE and PSY-FLAVOUR the most. In the second phase, despite the same amount of added onion flavour, onion odour and flavour intensities were highest in CTRL-FLAVOUR (6.6±1.4; 5.9±1.6), followed by PSY-FLAVOUR (5.7±1.1; 5.3±1.5) and NaA-FLAVOUR (4.6±2.6; 4.0±1.7). Hydrocolloids did not significantly influence overall flavour, saltiness, or fracturability. However, hardness was perceived highest for NaA-FLAVOUR (4.9±1.6), followed by PSY-FLAVOUR (3.3±1.4) and CTRL-FLAVOUR (2.7±1.7). CTRL-FLAVOUR was overall liked the most (7.6±1.2), followed by PSY-FLAVOUR (6.5±1.3) and NaA-FLAVOUR (5.5±1.6).

Conclusion: In addition to the flavouring technique, the ingredients that influence the release of the flavour should also be taken into account when developing the formulation. The addition of flavourings directly into the batter has proven to be beneficial for gluten-free 3D snacks.

Impact of roasting and coffee species on coffee powder characteristics and espresso coffee sensory liking

Dr Maria Di Cairano¹, Nazarena Cela², Teresa Scarpa¹, Nicola Condelli¹, Fernanda Galgano¹

¹School of Agricultural, Forest, Food, and Environmental Sciences; University of Basilicata, ²University of Gastronomic Sciences

Aim: This study aimed to investigate the effect of coffee bean species and roasting profile on chemical-physical parameters of coffee powder and consumer preferences for espresso coffee.

Method: Arabica (A) and Robusta (R) green coffee beans were roasted according to three roasting profiles: Full-City 210°C x 13' (FC), Italiana 220 °C x 14' (ITA), and Napoletana 230°C x 16' (NAP).

The roasted coffee powders were analysed for moisture content, colour (L* and chroma), solubility index, bulk density, total phenolic content, caffeine, and acrylamide content. For the sensory evaluation, 79 Italian consumers expressed their overall liking for each sample (9-point hedonic scale), and then they rated the intensity of colour, bitterness, acidity, sweetness, astringency, and characteristic flavour using a 5-point Just-About-Right (JAR) scale (1= too weak, 3= JAR, 5= too strong).

Results: As expected, Robusta coffees had higher ($p < 0.05$) caffeine content compared to Arabica (22.4 ± 5.00 vs 14.29 ± 0.29). Roasting profile significantly affected ($p < 0.05$) moisture content, L*, chroma values, and bulk density, with darker roasting profiles showing lower values for each variable. Though, total phenolic content exhibited an oscillatory trend, ITA samples had higher values than FC and NAP samples. Acrylamide content, ranging from 0.19 to 0.23 $\mu\text{g/g}$, resulted below the benchmark level set out in REG EU 2017/2158. Moreover, no significant impact of species and roasting profile on acrylamide content was observed.

Espresso coffees had liking scores comprised between 5.99 ± 1.74 (R-NAP) and 5.23 ± 1.73 (R-ITA). According to ANOVA results, coffee samples from darker roasting profile (NAP) were preferred by consumers, whereas species had no significant impact on coffee liking. Penalty analysis, conducted on JAR data to identify the drivers of liking, showed that consumers penalized espresso coffee when the perceived bitter, sour and astringent intensity was *too strong*, and the sweetness and characteristic flavour intensity was *too weak*.

Conclusion: The preference for darker roasted coffee samples aligns with the traditional Italian coffee drinking practices, particularly in southern regions. Moreover, this study identified the main sensory attributes penalising coffee liking, providing guidance for a proper selection of coffee species and roasting profile. This information could help coffee producers tailoring production process to consumer preferences.

Impact of smoking on quality characteristics of meat analogues compared to meat sausages

Dr. Ing. Olivier Goemaere¹, Prof. dr. ir. Ilse Fraeye¹, Annabel Meyfroot¹, Ing. Jolien Devaere¹, Ing. Ann De Winne¹, Dr. Ir. Myriam Loeffler¹

¹KULeuven

Aim:

Despite the wide availability of smoked meat analogues, their quality often falls short, particularly in flavour and colour (stability). Surprisingly, scientific literature on smoking of meat analogues is non-existent. To address this gap, this research aims to compare the quality aspects of a smoked emulsified meat analogue with that of a meat-containing cooked sausage using an identical smoking procedure.

Method:

Meat-containing sausage, serving as the quality bench mark, was processed by grinding lean muscle tissue, pork backfat, ice, kappa-carrageenan and spices in a bowl chopper. Key ingredients for the meat analogue included soy protein isolate (similar amino acid profile as the applied animal protein), sunflower oil, ice, kappa-carrageenan and spices. Both batters were stuffed in cellulose casings and subjected to a heating and smoking process using liquid smoke atomization. To minimize variables, identical proximate composition (protein, fat and water content) and processing parameters were maintained for both products, except for a higher final heating temperature for plant protein gelation (94°C vs. 75°C for meat-containing sausages). Smoked and unsmoked products were evaluated in terms of pH, cooking/drying loss, dry matter content, texture, color and aroma profiling through HS-SPME-GC-MS analysis.

Results:

Overall, smoking had a similar impact on the quality of both meat sausages and analogues. It led to increased moisture loss during processing, small pH reduction, color alteration (L*-value decreased while a*- and b*-value increased), increased hardness and the introduction of a smoky flavour profile. Notably, no significant differences related to total amount of smoke volatiles were observed between the two matrices. Differences in product parameters between both smoked matrices (e.g., color, texture) were mainly attributable to pre-existing differences in the non-smoked versions.

Conclusion:

Smoking equally affects key product parameters of meat sausages and analogues with similar matrix composition, resulting in equally high-quality products. Further research is required to confirm whether the lower quality of commercially available smoked meat analogues may be attributed to lower protein content, different amino acid compositions, and/or increased hydrocolloid content. Nonetheless, smoked meat analogues with identical proximate composition show promise as a viable alternative to meat-containing smoked sausages.

Formulation of starch-based gels for dysphagic people: link between printability, rheological properties, and sensory characteristics

Dr Taise Toniazzo^{1,2}, Dr Cassandre Leverrier¹, Dr Giana Almeida¹, Prof. Carmen Cecília Tadini², **Dr Valérie Guénard-lampron**¹

¹Université Paris-Saclay, INRAE, AgroParisTech, UMR SayFood, ²Universidade de São Paulo, Escola Politécnica, Dept. of Chemical Eng., FoRC/NAPAN – Food Research Center

Aim:

Dysphagia is a disorder that has a negative impact on people's health and quality of life, as it directly affects the ability to chew and swallow food. 3D printing technology can contribute to the development of personalized textured food and healthy food products. Sensory analysis is a very useful tool for describing consumer perceptions and meeting their expectations and needs. In this study, starch-based gels were developed for people suffering from dysphagia, and their printability, sensory and rheological characterization were evaluated.

Method:

The starch-based gels were produced using wheat, waxy, maize, or tapioca starches, and kappa-carrageenan (κ C) was used as a gelling agent. The starch-based gels were evaluated through a recovery test, which occurred in three different stages: (1) storage and loss moduli (G' , G'') were recorded at rest at a constant frequency (1 Hz) and strain (0.1%) for 120 s, (2) then, a constant shear of 10 1/s was applied for 40 s and, finally, (3) the first stage was repeated to assess structural regeneration. 3D food printing quality was evaluated of whether the starch-based gels were extruded and supported the layers deposited above it, without collapsing. Sensory analysis was carried out on printable products, with 12 panellists to measure the perceived intensity of 7 texture descriptors. Rheology and 3D printing were performed at 20 °C on the gels production day and sensory analysis was performed the following day.

Results:

Among the results obtained, we observed that the wheat starch-based gel was printable and obtained the desired shape after deposition. In contrast, the tapioca starch-based gel was not printable. These differences could be explained by rheological behaviour. The wheat starch-based gel showed a recovery of G' of $(87.22 \pm 2.92) \%$ and G'' of $(89.82 \pm 2.08) \%$. Conversely, the tapioca starch-based gel showed a G' recovery of $(65.02 \pm 3.32) \%$ and a G'' of $(83.16 \pm 1.42) \%$.

Conclusion:

The rheological measurements were correlated with printability and sensory characteristics such as firmness, adhesiveness, and swallowing difficulty of the printable starch-based gels. These results can provide a useful tool for formulating products suitable for dysphagic people.

Sensory characterization of infant cereals: A case study of reformulated recipes

Dr. Michelle Klerks¹, Dr. Maria Jose Bernal¹, Victoria de Lucas¹, Dr. Luisma Sanchez-Siles¹

¹Hero Institute for Nutrition, Hero Group

Aim: Infant cereals are common complementary foods. Some commercially available infant cereals still contain fruit juice concentrates. Such concentrates are generally used to provide a fruity flavor, but also provide sweetness and are considered free sugars. Manufacturers are encouraged to reformulate their products towards healthier and more natural products by removing fruit juice concentrates while mitigating significant alterations to sensory attributes. This study tested how an improvement in the recipes of infant cereals (gruel and porridge), by substituting fruit juice concentrates with fruit purees, would impact their sensory profiles.

Method: Three infant cereals pairs (original *versus* reformulated recipes of one gruel and two porridges) differing in flavor and target age were tested. Main differences between the original and reformulated recipes were the replacement of fruit concentrates by fruit purees. A total of 22 and 20 semi-trained panelists assessed one gruel and two porridge pairs, respectively. Samples were monadically presented in random order and were evaluated using the Rate-All-That-Apply method. Panelists received a list of taste and flavor attributes (sweetness, cereal flavor, fruitiness, milkiness) and texture attributes (homogeneity, creaminess, viscosity). They were instructed to choose the applicable attributes for each sample and rate their intensity on a 9-point scale, ranging from *low* to *high*. Paired T-tests were used to test for differences among each pair of infant cereals.

Results: Among all three pairs of infant cereals, no significant differences were observed between the original and reformulated recipes for taste and flavor attributes. The sensory profiles of the original and reformulated gruel recipes were perceived very similar, with only a minor significant difference in homogeneity. The reformulated recipe of one porridge pair demonstrated a slight but significant increase in homogeneity and creaminess, whereas the reformulated recipe of the other porridge pair exhibited decreased homogeneity and creaminess but increased viscosity, as compared to the original recipes.

Conclusion:

These findings suggest that a reduction in free sugar by substituting fruit juice concentrates with fruit purees in infant cereals does not significantly alter taste and flavor perceptions, which may encourage manufacturers to reformulate their infant cereals towards healthier and more natural products.

The importance of sensory evaluation in the development of polysaccharide-based edible gels for the elderly

Melina Korčok¹, Radoslav Židek¹, Vladimír Vietoris¹

¹Institute of Food Sciences, Faculty of Biotechnology and Food Sciences, Slovak University of Agriculture in Nitra

Aim: Addressing the complex issues of maintaining optimal health and nutrition is extremely important and often poses challenges for specific demographic groups. Older people in particular face a spectrum of physiological and pathological changes that significantly affect their eating habits and overall well-being. In this context, the development of easy-to-eat foods that provide certain nutritional benefits is essential. The objective of this study was to develop polysaccharide-based edible gels for the elderly population and to conduct consumer tests to identify specific factors and sensory properties that are critical for consumer preferences.

Method: Three phases of sensory evaluation involving seniors aged 65 years and over (N=80) were conducted, with different flavors of beta-glucan (BG) and arabinogalactan (AG)-based edible gels used in each phase. These flavors included: cocoa and vanilla, fruit, and coffee flavors. A 9-point hedonic scale was used to evaluate attributes such as appearance, aroma, flavor, aftertaste, and overall liking. The Just About Right (JAR) scale was used to assess the fluidity, color, intensity of sweet taste, intensity of bitter/sour taste, and intensity of flavor.

Results: The flavor of edible gels significantly influenced the sensory preferences of older consumers, with apple crush, coffee cake, and latte being the most preferred options. In addition, differences were observed in the evaluation of samples containing two different polysaccharides, indicating that the evaluators preferred the AG-based edible gels. The attributes evaluated on a 9-point hedonic scale represent the principal attributes for consumer evaluation of edible gels, with statistically significant differences found at each phase at the $\alpha = 0.05$ level of significance. The intensity of flavor was identified as a key attribute affecting the overall liking of samples at all three stages of the research.

Conclusion: Edible gels represent a promising food form for seniors, with consumer acceptability largely dependent on product flavors. Sensory evaluation can be a useful method in new product development for the optimization of a product based on consumer preferences.

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Texturing as structure enhancer of strawberry chips to promote healthy snack consumption

Msc Demian Martini-loesch¹, Joy Chidinma Nnadiogbulam¹, Annika Kofler¹, Michael Gasser¹, Massimo Zago¹, Elena Venir¹

¹Laimburg Research Centre

Aim:

The project focused on developing a healthy, additive-free snack to valorize the strawberries cultivated in various valleys of Alto Adige, Italy.

Method:

Strawberries purchased from the local market were washed, sliced and subjected to swell drying using instant controlled pressure drop (DIC) technology. This process is a high-temperature, short-time treatment followed by decompression through a rapid (instantaneous) exposure to vacuum. The decompression induces self-evaporation, resulting in tissue expansion (texturing) and yielding a distinctive consistency and porosity, thereby reducing the overall drying time. A combination of three drying steps was applied: pre-drying at low temperature with a heat pump drier, texturing through a prototype pilot plant (DIC), and post-drying under vacuum. Different processing parameters (initial slice thickness, drying times and temperatures) were tested to determine the optimal settings that yield the best expansion and lowest water activity in the final product. Three strawberry varieties; Elsanta, Falco, and Aprica were selected for the final product due to their agronomic and functional properties. Locally produced strawberries from these varieties were washed, sliced and dried according to predefined settings based on the preliminary results, including and excluding the DIC step. The water activity (*aw*), colour, expansion and glass transition temperature of the resulting products were then measured and compared.

Results:

Best settings for optimal product quality and physical properties were established as a slice thickness of 10 mm, pre-drying for 22h at 35°C, texturing at 150 C for 7 sec and 85 mbar and post drying for 4h at 60°C. The average total color differences (ΔE) between DIC-treated and non-DIC treated strawberry slices of Elsanta, Falco, and Aprica were 9.22, 7.84, and 10.68, respectively. The glass transition temperature of DIC treated samples was 2°C higher than the non DIC treated samples at an adjusted *aw* 0.33.

Conclusion:

As anticipated, the DIC treatment significantly reduced the *aw* value, resulting in the desired crispy texture. Data on chemical-physical and sensory quality are discussed. By exploiting DIC technology, this research contributes to the development of a healthy and flavorful snack, while valorizing locally grown strawberries in Alto Adige, Italy.

Rate-all-that-apply sensory evaluation in the characterization of cookies with pumpkin powder

Milana Matic¹, Nikola Maravić¹, Dragana Ubiparip Samek¹, Nataša Đerić Ilić¹, Alena Stupar¹, Anamarija Mandić¹, Mladenka Pestorić¹, Jelena Tomić¹

¹Institute of Food Technology (FINS)

Aim: The research aimed to sensory characterization of cookies made with the addition of pumpkin powder using two different types of fats. The cookie formulations included 30% pumpkin powder combined with either vegetable fat or butter. To further differentiate the cookie profiles, the evaluation also included a commercial cookie sample with a composition similar to that of the tested samples.

Method: Sensory attributes collected from the literature were selected through Rate-All-That-Apply (RATA) applied to five types of cookies. Sensory testing was conducted at the accredited sensory lab (Institute of Food Technology, Novi Sad) under controlled environmental conditions in individual, fully equipped sensory booths. The evaluation was performed by a semi-trained panel of 15 assessors. Assessors were instructed to select from a list of descriptors all sensations that described the samples and to rate their intensity on a 5-point scale.

Results: A list of 46 sensory attributes was compiled for the characterization of individual cookie samples. A two-way ANOVA model (product and replicate) was applied to RATA intensity scores, focusing only on attributes already selected for the Venn diagram. Tukey's post hoc test was used to determine differences between cookies, with significance set at $P < 0.05$. Mean RATA intensity values were utilized to create spider plots representing the sensory profiles of each cookie.

Conclusion: This research showed that RATA can serve as a reliable alternative to traditional vocabulary development methods, not only in selecting sensory descriptors but also in highlighting their significance. It provides insights into the sensory variability of cookies made with pumpkin powder and suggests its potential utility as a sensory tool for further cookie development and commercialization.

Effects of exopolysaccharides on the sensory quality of fermented dairy products

María Fernanda Alba Pinto¹, Bertha Viviana Ruales Guzmán²

¹Universidad Nacional De Colombia, Instituto de Ciencia y Tecnología de Alimentos, ²Universidad Nacional De Colombia, Instituto de Ciencia y Tecnología de Alimentos

Aim:

Most dairy products have a variety of additives within their ingredients to improve their sensory properties. However, current market trends demand products with less inclusion of additives in their matrix. As a result, the aim of this work was identified studies associated with the effects of exopolysaccharides on the sensory quality of fermented dairy products as substitutes of additives.

Method:

To achieve the aim of this work, a systematic literature review was used as methodology and Scopus as a database, to identify the literature related to effects of exopolysaccharides on the sensory quality of fermented dairy products such as alternative for the replacement of some additives in this type of food.

Results:

According to the systematic literature review, exopolysaccharides are biopolymers produced, mainly by microorganisms such as *Lactococcus*, *Lactobacillus*, *Leuconostoc*, *Pediococcus* and *Streptococcus thermophilus*, which are used for milk fermentation. The interaction between exopolysaccharides and milk proteins is enhanced by the high molecular weight of biopolymers and the configuration and distribution of their monomers, which can bind to the protein through hydrogen bonds and form the protein-exopolysaccharide complex. The presence of this complex affects the physico-chemical properties of the product, such as viscosity, water retention capacity, firmness, cohesion and particle size. Therefore, the incidence of exopolysaccharides on the above properties lead to improve the sensory characteristics of dairy products related to texture, mouthfeel, appearance and accentuate the typical flavors of this type of products.

Conclusion:

The presence of exopolysaccharides in dairy products would be an alternative to replace some additives normally used in these foods, since they allow to improve some of their sensory properties through the formation of the protein-exopolysaccharide complex. Furthermore, several authors suggest the combination of strains (commercial and modified) increase the efficiency of exopolysaccharides production, suggesting these polymers as substitutes for thickeners and emulsifiers.

Development of functional gluten-free breads adapted to the nutritional requirements of coeliac patients

Jhazmin Quizhper¹, Pablo Ayuso¹, Rocio Peñalver¹, M.A Rosell¹, **Professor Gema Nieto**¹

¹Universidad de Murcia

Development of functional gluten-free sourdough breads with antioxidant extracts and dietary fibre adapted to the nutritional requirements of coeliac patients.

Aim: The aim of this study was to formulate gluten-free sourdough breads improving the nutritional value of gluten free breads on the market, using broccoli extracts obtained from food industry by-products and enriching them with fibre.

Method: Two gluten-free sourdough breads were made with two different broccoli extracts: one with and one without enzyme treatment, both enriched with dietary fibre using flax seeds and flour from quinoa, chickpea and buckwheat. A commercially available gluten-free bread was used as a control. Nutritional composition, dietary fibre, antioxidant capacity (FRAP, DPPH and ABTS) and total phenolic compounds (Folin), colour, pH and sensory analysis were studied to evaluate the organoleptic quality of the different samples compared to the commercial bread.

Results: The results showed that the incorporation of flax, broccoli extract and quinoa, chickpea, and buckwheat flours increased the percentage of total dietary fibre. In addition, the two reformulated breads also showed significantly improved antioxidant properties compared to commercial bread. On the other hand, the enzyme-treated bread showed the best results in total dietary fibre (13.65%), total phenolic compounds (461.36 mg gallic/g) and antioxidant capacity tests. In addition, in the sensory analysis, the bread with enzyme-treated extract was rated higher than the bread without enzyme treatment.

Conclusion: In conclusion, taking into account the nutritional, physicochemical and organoleptic characteristics, broccoli extract with enzymatic treatment is the ideal compound for the production of fortified sourdough breads for people with coeliac disease.

Sensory drivers of liking and consumer emotional responses to vegan fish vs fish

Marta Appiani¹, Camilla Cattaneo¹, Monica Laureati¹

¹ Sensory & Consumer Science Lab (SCS_Lab), Department of Food, Environmental and Nutritional Sciences (DeFENS), University of Milan, 20133 Milan

Aim:

Today, the demand for alternative seafood is rapidly increasing. Plant-based breaded fish fillets and fingers are highly popular imitations, yet little is known about consumer experiences with these products. This study aimed to: a) explore sensory, liking, and emotional responses to commercially available plant-based breaded fish alternatives; and b) categorize consumers using an innovative clustering method based on cata and liking data (CLUSCATA-liking).

Method:

The present research explored plant-based/animal-based breaded fish product category using a multi-variable research strategy where 104 consumers (52% Female; 27.7±8.9 y.o.) tasted samples for degree of liking/disliking and described their perceived sensory characteristics using check-all-that-apply (CATA) questions. A product-specific questionnaire was developed to evaluate the emotional response associated with the product consumption.

Results:

GLM results showed a significant effect of samples on overall liking ($F=22.0$; $p<0.0001$). Although animal-based samples were the most liked, samples formulated with dehydrated rice flakes, rice or texturised wheat protein were well accepted (Ls-mean=53.6-62.1). In contrast, samples made with soy and rice protein were disliked (<50) due to perceived bitterness, legume and vegetable odour and off-flavours. The differences among the samples were further emphasized based on the associated emotions. Animal-based samples were associated with emotions of satisfaction and happiness, while plant-based samples elicited negative emotions such as disappointment, disgust and indifference. Only the sample containing textured wheat protein evoked positive emotions (surprise, curiosity, novelty). Preliminary consumer segmentation using Cluscata-liking identified two distinct clusters with differing preferences for plant-based samples. Further analyses are in progress to characterise them in terms of the sensory and emotional responses.

Conclusion:

These findings show that incorporating the measurement of implicit variables such as emotions alongside liking leads to enhanced product differentiation. Through the multi-response approach adopted in this study, it was possible to obtain a comprehensive product characterisation, contributing to a better understanding of the factors driving the liking of plant-based fish alternatives, valuable for product development and marketing strategies.

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From omnivores to vegans: Understanding the Protein Transition Across Dietary Groups in Spain

Phd Ana Baranda¹, PhD n Da Quinta¹, Aniol Peracaula¹, Elena Santa Cruz¹, Sofia Roca¹

¹Azti

Aim:

In a social context in which people are more aware than ever of the environmental and social problems of our time, choosing food products that are beneficial for health and the planet is increasingly common. Accompanying the so-called protein transition, the increasing incorporation of ingredients in food products from protein sources other than the traditional one of animal origin is a reality. This transition is often not a sudden shift but rather a series of incremental changes. Consumers take small steps toward specific goals, adapting their eating habits based on encountered challenges. Understanding the personal motivations driving this transition provides valuable insights for product development and market orientation. The knowledge, attitude and barriers towards the different sources of alternative protein, as well as the demand for the type of products by the consumer or the expected place of purchase, represents not only key information for the development of new products that meet the demands of today's consumer but also for their future success.

Method:

After a qualitative approach in order to design the questionnaire and to increase the knowledge about the targets, a cross-sectional survey was conducted in Spain in 2022 and 2024 involving more than 1500 participants from different dietary patterns (same quota ranging from omnivores to vegans). The evolution of the different dietary groups differing in the level of animal food restriction regarding different aspects related to alternative protein was observed.

Results:

Our results show the evolution in recent years food trends and habits for each dietary group: reasons for adherence to the diet, time in which the transition was made, future transition, difficulties in following their eating style, perception and attitude towards the different sources of alternative proteins, barriers and enablers towards products made with those ingredients, or other aspects such as purchase expectations and where to find them.

Conclusion:

The results of this study will serve to identify new opportunities in the launch of products on the market in accordance with the expectations and needs of the consumer of the future.

Therapeutic potential of isolates from gamma irradiated onion skin against Breast cancer and Macrophage cells

Ms. Kavita Thukran^{1,2}, Dr. Vanshika Adiani^{1,2}, Dr. Deepak Sharma^{1,2}, Dr. Bibhuti Bhusan Mishra^{1,2}

¹Bhabha Atomic Research Centre, ²Homi Bhabha National Institute

Aim:

The outer skin of onion (OOS) is a waste material which could be a potential source of bioactive compounds with diverse applications in food, pharmaceutical, and cosmetic industries. The present study was aimed to develop a method for fractionation of bioactives and explore cytotoxic effects on human breast cancer (4T1) cells and macrophages (RAW 264.7) for effective utilization of OOS.

Method:

The OOS was gamma irradiated (10-25 kGy) using a Co⁶⁰ source and used for extraction of various bioactive compounds. The irradiated and non-irradiated onion skin powder was then fractionated into oil extract (using hexane), phenolics-rich extract (using ethyl acetate), and residual fibres (using acid and alkali hydrolysis). The phenolics-rich extract was characterized using Thin Layer Chromatography (TLC), Fourier Transform Infrared Spectroscopy (FT-IR) and Gas-Chromatography Mass spectrometry (GC-MS). The antioxidant and anti-inflammatory activity were estimated using standard *in-vitro* assays. The anti-cancer activity was evaluated using breast cancer and macrophage cell lines through MTT, clonogenic, and fluorimetric assays to analyze the cell viability, cell proliferation, and oxidative stress, respectively. Additionally, flow cytometry was used to assess apoptosis induction in 4T1 and RAW 264.7 cells using a PI staining protocol.

Results:

The OOS with absorbed dose of 10 kGy showed higher oil content (155±10%) and phenolics (35±2%), which could be due to increased extractability. The TLC, FTIR and GC-MS data showed quercetin as major (95%) phenolics among 14 different phenolic acids and showed increased content in 10 kGy treated OOS. Similarly, 10 kGy irradiated OOS showed 18±1.5% increase in antioxidant activity as compared to non-irradiated sample. The phenolic extract showed dose-dependent cytotoxicity against 4T1 and RAW 264.7 cell lines and proliferative activity in normal splenocytes. The clonogenic assay also demonstrated dose dependent anti-proliferative activity in both cell lines. Furthermore, flow cytometry results showed dose dependent increase in apoptotic cells. The results of this study revealed that phenolics-rich extract from OOS possesses potent antitumor and antioxidant properties.

Conclusion:

The 10 kGy irradiated OOS found to be a better source for isolation of bioactive compounds. The phenolics-rich extract from OOS possesses potent antitumor properties against studied breast cancer and macrophage cells.

Rice Bran: A Resourceful By-Product for Sustainable Food Innovation

Danilo Candela¹, Cecilia Fiore¹, Silvia Fraterrigo Garofalo¹, Emmanuele Parisi¹, Elena Simone¹

¹Politecnico di Torino

Aim:

In 2020, the global production of rice reached 758 million tons (FAO): its demand is expected to remain strong, leading to an increased generation of by-products. This study aims to valorise rice bran, a residue of rice production, by extracting and separating its lipid fractions, including high value compounds such as waxes. This work seeks to find the optimal extraction method to recover rice bran butter (RBB) and its fractions from rice bran. Additionally, rice bran wax (RBX) was tested for its potential as ingredient in innovative food formulations such as oleogel.

Method:

RBB was extracted from rice bran using supercritical CO₂, an efficient and environmental-friendly method. Chemical characterization techniques were used to determine the fatty acid (FA) and triacylglyceride (TAG) profile of the resulting butter. Subsequently, RBB was subjected to a series of different unit operations such as centrifugation, crystallization, and filtration to isolate two primary fractions: rice bran oil (RBO) and RBX. Differential Scanning Calorimetry (DSC) was used to evaluate the optimal purification method. Further structural analysis of the fractions involved synchrotron X-ray radiation scattering to identify and characterize the polymorphs present in solid RBX. The purest RBX fractions obtained were then used to produce oleogels with sunflower oil. Structural analysis, combining X-ray synchrotron radiation and rheology, was conducted to study the process of gel formation.

Results:

FAs characterization revealed a mostly unsaturated composition (~ 80 %w/w), with oleic acid being the major component (~ 50 %w/w of unsaturated content). The purification method with the most centrifugation steps achieved the highest purity and yield using isopropanol as solvent. The structural characterization of RBX indicated the prevalence of an orthorhombic β' sub-cell polymorph associated to the presence of policosanols. When RBX was used to produce oleogels, it was found that minimising shear stresses during gelation resulted in oleogels with a firmer texture and improved stability.

Conclusions:

The RBB and RBX extraction from rice bran represents a significant step towards sustainable food innovation. By improving food quality, this approach meets the growing demand for rice while paving the path for environmentally conscious solutions in the food industry.

Modulating structure and texture of heat and acid-induced gels by controlling the pH of milk

PhD candidate Zhe Cheng¹, Lilia Ahrné¹, Matthias Eisner², Pauline Leusden²

¹Copenhagen University, ²Yili Innovation Center Europe

Aim:

Paneer, a fresh, unripened cheese produced through the heat-acid coagulation of milk, stands out due to its unique characteristics. In contrast to rennet-induced cheeses, paneer maintains its structural integrity without melting upon heating, positioning it as a promising alternative protein source in the shift towards dairy-based meat substitutes. However, limited studies have explored the heat and acid-induced gelation as function of pH at which heat treatment is performed. Given the documented influence of pH on final composition of heated milk, the objective of this work was to study the effect of pH-adjustment of milk prior to heat-treatment on the subsequent gelation, and consequences for compositional, structural and textural properties of the heat and acid-induced milk gels.

Method:

Heat and acid-induced milk gels were produced by adjusting the pH (6.9, 6.7, 6.55, 6.35, 6.1) prior to heat treatment (90 °C for 12 min), followed by acidification to pH 5.2, then separation of the formed aggregates from serum. The effect of pH adjustment of milk on the compositional, structural and textural properties of the heat and acid-induced milk gels was studied.

Results:

Differences in the structural properties were observed among milk curds and the subsequent milk gels depending on the pH of milk prior to heat treatment. An increase in pH from unadjusted milk (pH 6.7) to 6.9 led to a significant reduction in particle size of the formed curds. Compared to unadjusted heat and acid-induced milk gels, those treated at pH 6.9 exhibited higher moisture levels, resulting in a higher yield with relatively lower protein and fat content. The results obtained for protein-protein interactions within the heat and acid-induced milk gels revealed that electrostatic interactions and hydrogen bridges (25-42%), and calcium bridges (30-45%), were the dominant protein interactions in all the gels. Heating milk at pH 6.7 and below caused a clearly increased amount of calcium-protein interactions compared to pH 6.9, yielding a more rigid and less hydrated gel, which aligns with compositional and image analyses.

Conclusion:

Variations in the pH value before milk heating impacted the physical properties of coagulum-serum mixture, and the structural features of the resulting gels.

Utilizing *Tinospora crispa* as a Functional Food Resource: Bioactive Compound Isolation via Supercritical Fluid Extraction

Hae Na Chi¹, Dongyup Hahn^{1,2}, Seongdo Lee², Seunguk Yu²

¹Department of Integrative Biology, Kyungpook National University, ²School of Food Science and Biotechnology, College of Agriculture and Life Sciences, Kyungpook National University

Aim:

This study focuses on exploring the bioactive substances in the supercritical fluid extracts of *Tinospora crispa*, which have been scarcely reported. *T. crispa*, is widely incorporated into traditional culinary practices across Southeast Asia, where its stems, leaves, and other parts are added to various dishes. Traditionally used for various medicinal purposes including diabetes, inflammation, and cancers, this inclusion in the diet is driven by the plant's acclaimed health benefits, underscoring its importance not merely in medicinal applications but also as a nutritious component of daily diets. The use of *Tinospora crispa* as a food item capitalizes on its potential therapeutic attributes, embedding it within the culinary traditions of the indigenous communities where it is found. This research seeks to identify and analyse structures of its components to better understand the therapeutic potentials

Method:

Supercritical fluid extracts of *T. crispa* was performed, followed by separation into hexane, dichloromethane, ethyl acetate, butanol, and aqueous soluble fractions. The hexane fraction was partitioned using medium pressure liquid chromatography (MPLC), while the ethyl acetate fraction was further subdivided by size exclusion chromatography. Compounds isolated from these layers were purified using preparative HPLC. Structural identification was conducted using NMR spectroscopy and LC/ESI/MS.

Results:

The study successfully isolated various compounds, including phenolic amides, from the hexane and ethyl acetate fractions. The anti-inflammatory potential of the compounds was evaluated by measuring their ability to inhibit lipopolysaccharide (LPS) induced nitric oxide (NO) production in BV2 cells. In addition, the cell viability of BV2 cells was determined using Cell Counting Kit (CCK)-8.

Conclusion:

The fractionation of *T. crispa*'s supercritical fluid extracts yields bioactive compounds with potential anti-inflammatory benefits. These findings not only enhance the pharmacological understanding of *T. crispa* but also support its traditional uses and pave the way for the development of functional foods or novel therapeutics based on *T. crispa*. Furthermore, this study suggests the potential of *T. crispa* as a functional material and indicates that phenolic amides could be used as marker substances in the development of high-value functional materials using *T. crispa*.

Curcumin Solid Particles for Enhanced Stabilization of Pickering Emulsions

Ms Giulia Del Duca¹, Professor Elena Simone¹, Dr. Fiора Artusio¹, Dr Eleonora Calì¹

¹Politecnico di Torino

Aim

The application of solid particles to stabilize foams and emulsions through the Pickering effect has emerged as a promising approach in food and pharmaceutical science formulations. This approach offers many advantages, including enhanced stability and prolonged shelf life. Moreover, their potential as effective encapsulation systems has attracted significant interest. In line with the growing demand for natural-derived compounds with desirable properties, such as health benefits, safety, and cost-effectiveness we chose curcumin, well-known for its antioxidant and antimicrobial properties. Curcumin also exhibits fascinating interface properties that make it an ideal candidate for food applications. Our study focuses on the utilization of submicron-sized curcumin particles as stabilizers, employing a crystal engineering approach to tailor particle properties.

Method

The submicron curcumin particles were obtained using the anti-solvent precipitation technique, using a mixture of water and ethanol. Additionally, we explored the influence of incorporating polymeric hydrocolloids—such as carboxymethylcellulose (CMC), chitosan, and alginate—into the aqueous phase, investigating their impact on curcumin particle surface properties. The particles obtained were tested as Pickering stabilizers to stabilize emulsions.

Results

Curcumin particles show a clear affinity for stabilizing water-in-oil (W/O) emulsions. The attachment of curcumin particles at the interface between the two phases was observed, resulting in a stabilization of water droplets. This strong interaction results in effective stabilization at the water-oil interface, demonstrating the potential of curcumin as a promising Pickering stabilizer for W/O emulsions, preventing phase separation and maintaining a well-dispersed system.

Conclusions

By elucidating the relationship between crystal properties and their role as stabilizers in Pickering emulsions, our study contributes to expanding the knowledge in this field, paving the way for the formulation of innovative food products.

Innovative solution for the replacement of methylcellulose in plant-based burgers

Food Science Engineer Julie Deviers¹, Zoé PALE CZNY², Sabine PHOMPHOUSA², Maëlys SAINT-LOUIS², Laurent Lethuaut^{2,3}, Lizeth LOPEZ¹

¹Mane, ²ONIRIS National College of Veterinary Medicine Food Science and Engineering, ³ONIRIS Nantes University CNRS GEPEA UMR 6144 MAPS² Team Flavour platform

Aim: In recent years, global consumption of plant-based meat alternatives has surged, yet concerns persist among some consumers regarding the perceived healthiness of certain ingredients, notably methylcellulose—a widely used binding agent in plant-based burgers valued for its unique texturizing properties. Recent research suggests that combining pectins with enzymes could offer a promising alternative to methylcellulose. This study seeks to identify the optimal formulation for replacing methylcellulose in plant-based burgers using the synergy between laccase, sugar beet pectin (SBP) and faba protein isolate (FPI).

Method: An experimental design assessed the effects of laccase, SBP, and faba protein isolate (FPI) on plant-based burger texture across 17 products, evaluated by a trained sensory panel (n=16). Multi-response optimization determined the concentrations of laccase, SBP, and FPI that optimized firmness, juiciness, and stickiness. Concurrently, Texture Profile Analysis (TPA) and juice release measurement provided instrumental insights. Partial least square (PLS) regression identified correlations between sensory and instrumental data.

Results: Multi-response optimization yielded a formulation with a desirability index of 0.94, aligning with sensory attributes akin to methylcellulose-based burgers (target). PLS regression revealed correlations between sensory and analytical parameters, enhancing our understanding of plant-based burger texture and formulating strategies for improvement.

Conclusion: This study displays the feasibility of replacing methylcellulose in plant-based burgers with an optimized formulation leveraging laccase, SBP & FPI. Achieving a desirability index comparable to methylcellulose-based burgers underscores the potential for enhancing the perception and acceptance of plant-based meat alternatives.

The storage stability and *in vitro* digestion of flaxseed oil bodies with different interfacial molecules

Rao Fu¹, Yu Peng, Mo Li, Yuanying Ni, Remko Boom, Costas Nikiforidis, Xin Wen

¹China Agricultural University

Aim: Flaxseed oil bodies (FOBs) are the intracellular vesicles that store oils within flaxseeds. By employing specific aqueous isolation procedures, FOBs with diverse interfacial molecular compositions can be produced. Consequently, the unique molecular makeup of the FOB interfaces may significantly influence their storage stability and digestive properties. These characteristics are paramount considering the potential use of FOBs in applications such as fat substitutes or systems for delivering bioactive compounds.

Method: Crude FOBs (CFOB) were obtained from flaxseeds using an enzyme-assisted aqueous extraction method, which were further purified by washing with pH 11.0 alkaline solution to produce the purified FOBs (PFOB). Natural FOBs (NFOB) were extracted aqueously without enzyme pretreatment. The microstructure and zeta potential of the FOB emulsions were monitored throughout 18 days' storage at 4°C. In addition, changes in microstructure, average particle size, zeta potential, protein composition, and free fatty acid release rate of different FOB emulsions during *in vitro* digestion were determined.

Results: The interfacial molecules in FOBs were primarily composed of polysaccharides and proteins, with a significantly higher amount of polysaccharides in NFOB compared to other FOBs ($P < 0.05$). Both NFOB and CFOB contained extrinsic proteins and oleosins (16-18 kDa), but PFOB only had oleosins. During the 18-day storage period at 4°C, oil leakage and emulsion stratification were observed in PFOB and NFOB, respectively. In terms of *in vitro* digestion, aggregation of CFOB and PFOB was observed under gastric conditions, which might be due to the complete degradation of oleosins by pepsin. However, NFOB remained dispersed in the digestive fluid with droplet size below 3 µm. Upon transit to the intestinal phase, the minimum $D_{4,3}$ value (1.29 µm) and the highest FFA release percentage (44.6%) were observed in NFOB after two hours of digestion.

Conclusion: CFOB exhibited the best physical stability, which might be attributed to extrinsic proteins inhibiting the creaming of emulsion, while polysaccharides could accelerate the flocculation of emulsion. NFOB maintained the minimum droplet size in the gastric phase, but released the highest FFAs during intestinal digestion, suggesting that FOBs have great potential to be a co-delivery vehicle for bioactive compounds.

Effect of changes in salt (NaCl) concentration and pH on rapeseed protein solubility

Lisa Gotzmann¹, Marcos H. Vinde¹, Lilia Ahrné¹, Marianne N. Lund¹

¹Copenhagen University

Aim: The optimal conditions for solubilizing rapeseed proteins and the effect of changes in pH and the removal of salt on protein solubility under conditions that may occur in an electro dialysis (ED) process were investigated.

Rapeseed, rich in storage proteins cruciferin and napin, emerges as a promising plant-based protein alternative. For future isolation of rapeseed proteins, solubility is a critical characteristic. Changes in the NaCl concentration or the pH value of the medium affect the solubility of cruciferin and napin in different ways due to their inherent molecular differences. ED is an effective and sustainable way to demineralize solutions, and we hypothesize that the protein solubility decreases at low NaCl concentrations leading to protein precipitation during ED.

Method: Rapeseed protein powder (CanolaPro[®], DSM) was dispersed in varying NaCl concentrations (1 – 0 M) and pH values (8.5 – 4.0). By reducing the NaCl concentration and lowering the pH value, an ED process is mimicked. Protein content and composition of the supernatant were analyzed at different salt concentrations and pH values using the Dumas method, and the protein composition was analyzed by RP-HPLC after centrifugation.

Results: Cruciferin solubility was high at NaCl concentrations down to 0.1 M NaCl and decreased at lower NaCl concentrations for all pH values except pH 4.0. At pH 4.0, a reduction in NaCl concentration lower than 0.1 M resulted in higher cruciferin solubility. Napin solubility did not improve with NaCl removal by lowering pH from 8.5 to 6.5 but showed improved solubility at pH 4.0 for 1 M – 0.1 M NaCl.

Conclusion: ED salt removal simulation shows that the solubility of napin and cruciferin is affected differently by NaCl concentration and reduction in pH. Cruciferin becomes less soluble at low NaCl concentrations across all pH levels (8.5 – 6.5) while at pH 4.0, higher salt concentrations resulted in its loss from the supernatant. In contrast, napin solubility remains unchanged upon NaCl reduction at pH 8.5 – 6.5 but notably increases at pH 4.0 at 1 M – 0.1 M NaCl. The precipitate seen during desalting by ED can be explained by precipitation of mainly cruciferin at low NaCl concentration.

Exploring the nutraceutical and technological properties of taro corns grown in Cyprus

Dr Atalanti Christou¹, Mrs Evgenia Siameti², Mr Alexandros Antoniadis², Professor Christos Ritzoulis²,
Dr Vlasios Goulas¹

¹Cyprus University Of Technology, ²International Hellenic University, Department of Food Science and Technology, Alexander Campus,

Aim: Taro (*Colocasia esculenta* L.) is an edible plant with one or more starchy corms that is widely cultivated in Asia and Africa. Cyprus island is the unique taro producer in Europe; recently Cypriot taro corns were recognized as Protected Designation of Origin (PDO) product. They comprise a high amount of carbohydrates, a special protein composition, significant of vitamin C and B contents as well as a variety of phenolic compounds and carotenoids. The objective of this work was to evaluate the phytochemical composition and antioxidant properties of taro corns grown in Cyprus. In addition, the capability of taro corn to act as source of thickeners and/or oleosomes is to be explored.

Method: An ultrasound assisted extraction using hexane, acetone, methanol, and water was applied on taro corns. At first, total phenolic (TPC) and flavonoid (TFC) contents were determined. Then, phenolic composition of extracts was studied with the employment of liquid chromatography. The antioxidant potential of extracts was assessed using DPPH free radical scavenging activity and ferric reducing antioxidant power (FRAP) assays. At the same time, thickeners were isolated from taro corns, and were examined in terms of their colloidal properties, biopolymeric population composition and rheology. In addition, oleosomes have been extracted from the same source, and the relevant preliminary results are shown.

Results: TPC values ranged from 0.11 mg GAE g⁻¹ to 5.53 mg GAE g⁻¹, whereas TFCs were between 0.25 mg CE g⁻¹ and 3.26 mg CE g⁻¹. Chromatographic separations showed that corns contain significant amounts of epicatechin and hydroxybenzoic acid. Regarding their antioxidant activity, results demonstrated that methanolic extract had the highest potential, followed by aqueous, acetonetic and hexanic extract. The rheological behavior of the extracted polysaccharides is presented and discussed under the light of their colloidal properties, while an interesting and previously unreported discovery pertains to the successful extraction of oleosomes.

Conclusion: The present study demonstrates the bioactive composition and technological properties of PDO taro corns grown in Cyprus. They are a valuable source of small bioactive metabolites and potential source of hydrocolloid thickeners and oil bodies, with great importance for food industry.

Aerogel-Based Clarification of 'Ice' Tea

Ozge Guven¹, Serap Namli¹, Emre Taskin², Mecit Halil Oztop¹

¹Middle East Technical University Food Engineering Department, ²Doğadan Food Products Research and Development Department

Aim:

The main purpose of this study is to use chitosan and xanthan gum aerogels to naturally clarify cold brewed teas. This novel method aims to enhance the visual and sensory qualities of tea while preserving its bioactive characteristics and providing a cost-effective and environmentally responsible substitute for traditional methods.

Method:

In this study, aerogels made from chitosan and xanthan gum clarified black and green tea infusions. The method involved brewing, cooling, and treating with aerogels, then comparing their effectiveness to traditional silica adsorbents. Clarification impacts on turbidity, color change, and total solids were measured. Total solid content was determined by the sample weight ratio before and after drying at 105 °C. Color differences were calculated from the tea's lightness (L*), red/green (a*), and yellow/blue (b*) balance values. Polyphenolic content was analyzed by the Folin-Ciocalteu method, reported in mg GAE/g dry tea. Total protein content of the tea after clarification was measured by Bradford method.

Results:

The findings showed that aerogels, particularly in black tea, greatly decreased turbidity. In comparison to the conventional clarification by using silica, chitosan aerogel was shown to be more successful in terms of clarity. The aerogels' selective adsorption ability was demonstrated, despite the reported reductions in the total amount of polyphenols, there was a noticeable effect on the total solids for chitosan treated samples. Improvements in physical properties of tea such as color and clarity have made chitosan aerogels a potential adsorbent to use in clarification processes.

Conclusion:

The study suggests that the tea industry can use aerogels as efficient and sustainable clarifying agents. Using of aerogels as adsorbents would be an important progress for beverage industry since they have the potential to enhance the appearance and sensation of tea. To further improve adsorbent properties of aerogels by tuning polymer concentration, drying conditions etc., might help better clarification results and achieving the reusability of aerogels for the improvement of cost efficiency might be the future research to concentrate on.

Isolation and Bioactivity Evaluation of Phytochemical Compounds from *Salvia Hispanica* L. (Chia) Seed Oil Residue

Professor Dongyup Hahn¹, Hae Na Chi¹, Seunguk Yu¹, Dongyup Hahn¹

¹Kyungpook National University

Aim: The residue of *Salvia hispanica* L. (chia seed), obtained during the extraction of chia oil, is a by-product that remains underutilized. This study aims to investigate the phytochemical and bioactive properties of the chia seed oil residue to enhance its value as a product with high added value.

Method: Phytochemical extraction was performed using a dichloromethane/methanol (DCM/MeOH) mixture, followed by fractionation with hexane, ethyl acetate, butanol, and water. Fractions obtained with hexane and ethyl acetate were subjected to vacuum liquid chromatography to yield fractions HX1-HX4 and EA1-EA3, respectively. The EA1-EA3 fractions underwent further fractionation using Sephadex LH-20 size exclusion chromatography. Component compounds were purified by preparative high-performance liquid chromatography (HPLC), with their structures elucidated via liquid chromatography-mass spectrometry (LC-MS) and proton nuclear magnetic resonance (¹H-NMR). The immunomodulatory and antioxidant activities of these compounds were evaluated to assess their bioactivity.

Results: Four phenolic compounds were isolated and characterized. Utilizing these compounds as indicator components, a standardized extraction procedure was established, optimizing temperature, time, and solvent parameters to maximize the immunomodulatory and antioxidant effectiveness of the extracts.

Conclusion: A standardized method for isolating compounds from the residue of chia seed oil extraction has been developed. The potential of chia seed residue as a high-added-value product was confirmed through the evaluation of the immunomodulatory and antioxidant activities of the isolated compounds. Further development and testing, both in vitro and in vivo, are recommended to fully harness the high-added value of chia seed residue.

Preparation and characterization of protein-stabilized O/W emulsions with natural waxes

Seunghyeon Han¹, Suyong Lee¹

¹Department of Food Science & Biotechnology and Carbohydrate Bioproduct Research Center, Sejong University

Aim :

There is a growing focus on natural emulsifiers with increasing consumer interest in sustainable consumption and natural origin foods. Especially, O/W emulsions utilizing proteins as emulsifiers have garnered significant interest for food and beverage formulations. However, O/W emulsions can cause flocculation or oxidation of the oil due to the oil existing in a liquid state. Therefore, the objective of the study was to enhance the stability of protein-stabilized O/W emulsions by solidifying the oil component with natural waxes.

Method :

Three different natural waxes (candelilla wax, carnauba wax, and beeswax) were incorporated into the formulation of protein-stabilized O/W emulsions. The formation of O/W emulsions was confirmed using the confocal laser scanning microscope with Nile red dye and their rheological properties were analyzed using a controlled rheometer. Furthermore, their oil droplet sizes were measured.

Results :

The confocal laser scanning microscopic analysis showed that protein-stabilized O/W emulsions were successfully generated using the natural waxes. Furthermore, the particle size of the emulsions showed that the beeswax emulsion had the largest particle size, followed by carnauba wax, candelilla wax. The viscosity of the emulsions decreased as the shear rate increased, clearly exhibiting shear-thinning behavior. The viscoelastic properties of the emulsions exhibited a sequential enhancement, correlating with the viscosity trend, with candelilla wax demonstrating the highest, followed by carnauba wax, and then beeswax.

Conclusion :

This study compared the influence of different natural waxes on the physicochemical characteristics of protein-stabilized O/W emulsions. According to the analysis of rheological properties and particle size, it was confirmed that the emulsions with smaller particle sizes exhibited higher viscosity and viscoelasticity. The findings of this study are anticipated to offer valuable insights for food research applications involving natural emulsifiers and waxes.

Differences in interfacial alignment of common chocolate surfactants

Miss Charlotte Huddart¹, Joselio Vieira², Fotis Spyropoulos¹, Bettina Wolf¹

¹University of Birmingham, ²NPTC Confectionery - Nestle UK

Aim:

The flow properties of molten chocolate are influenced by processing conditions and differing formulations. Lecithin and polyglycerol polyricinoleate are common surfactants used by chocolate manufacturers to tailor the flowability; lecithin has a larger impact on the viscosity while polyglycerol polyricinoleate (PGPR) reduces the yield stress. PGPR is perceived negatively by consumers and there has long been the desire to replace PGPR with a natural alternative. However, the unique yield stress reducing functionality makes this extremely challenging. In view to overcoming this challenge, the overall aim of this research is to further understand the interrelation between the chemistry of these emulsifiers, their molecular organisation in cocoa butter, and the behaviour at the interface of the sugar particles.

Method:

The interaction between chocolate surfactants and sugar particles was assessed via interfacial property measurements at the oil-water interface. Sunflower oil with added lecithin or PGPR of up to 2 wt% was selected to represent the continuous phase of molten chocolate, and water to mimic the hydrophilic surface of sugar. Measurement temperature was varied between 20 – 60 °C.

Results:

With PGPR the interfacial tension and time taken to reach equilibrium decreased with increasing concentration. While equilibrium interfacial tension for lecithin also decreased, time to equilibrium was inversely correlated to concentration. Based on data obtained at varying temperature, a structure mechanistic model will be discussed to explain the observations, including a link to the functionality of either surfactant in chocolate.

Conclusion:

Analysis of the interfacial behaviour of PGRP and lecithin at a model chocolate interface allowed to develop a hypothesis for their respective functionality in chocolate. These insights inform molecular structure requirements for PGPR substitution.

Encapsulation by spray drying: The influence of maltodextrin properties on retention of hydrophobic volatile d-Limonene

Encapsulation By Spray Drying: The Influence Of Maltodextrin Properties On Retention Of Hydrophobic Volatile D-limonene Ana Jauhari¹, Patrick Wilms¹, Meinou Corstens¹, Maarten Schutyser¹

¹Laboratory of Food Process Engineering, Wageningen University and Research, P.O. Box 17, 6700 AA

Aim:

Volatile compounds in food significantly impact consumer perception, with solid ingredient forms preferred for ease of incorporation. Spray drying is a common method for encapsulation, crucial for ensuring high-quality ingredients by enhancing volatile retention during processing. The choice of wall material, typically maltodextrin, is paramount as it determines emulsion behavior during spray drying, influencing the final product's quality. Maltodextrin's varying dextrose equivalent (DE) values affect skin formation rate and final morphology. However, there is limited research on the influence of DE on hydrophobic volatile retention and its relationship with surface oil content. Therefore, this study aims to investigate the influence of DE on the surface oil and retention of d-Limonene during spray drying encapsulation.

Method:

In this study, d-Limonene was chosen as a model volatile compound and dispersed in stripped sunflower oil. Four different DE values of maltodextrin were used as wall material and pea protein isolate was used as an emulsifier. The composition of the initial emulsion and the drying conditions were kept constant throughout the experiment. The Buchi-Mini Spray Dryer was utilized to produce the powder, with the feed pre-homogenized via microfluidization. Analysis of the initial emulsion and spray-dried powder was conducted. The relationship between surface oil and d-Limonene retention on dried powder was specifically addressed.

Results:

The retention of d-Limonene in the spray-dried powder significantly decreased with decreasing DE-value of maltodextrin. While the powders exhibited uniform size distribution, those with lower DE values showed higher surface oil content. The morphology of the final powder – characterized by the presence of pores and cavities – appeared to play a more significant role in d-Limonene retention than differences in skin formation rates through viscosity.

Conclusion:

In summary, our study highlights the critical role of DE-value in retaining d-Limonene. Our findings showed the crucial relationship between surface oil content and particle morphology, demonstrating that controlling particle morphology is pivotal for improving the quality of spray-dried powder. Mitigating surface oil contributes not only to volatile retention during spray drying but will also contribute to better preservation of encapsulated hydrophobic volatiles during storage.

Interest of a new emerging technology (Electrostatic Spray drying) to stabilize micro-algae

Audrey Maudhuit², Elodie Beaupeux², Jean Maxime Edoorh², Preethi Jayaprakash^{1,2}, Pr Claire GAIANI¹, Stephane Desobry¹

¹Université de Lorraine (LIBio), FRANCE, ²Fluid Air

Aim: One part of the industrial FLUID AIR project is related to stabilization of micro-algae under powder using an emerging technology: Electrostatic Spray Drying (**ESD**). For this purpose, ESD efficiency was compared to classical drying techniques such as Spray Drying (**SD**) and Freeze Drying (**FD**). Both have been widely used in commercial industries for food, nutraceuticals and pharmaceutical applications. The aim of this work is to position a new technology in comparison with traditional technologies by focussing on sensitive bio-active molecules, here micro-algae.

Method: Two micro-algae were used (*Chlorella Vulgaris* and *Chlorella pyrenoidosa*) and dried with the three technologies (i.e. ESD, SD and FD). The obtained microparticles were evaluated based on their morphology, size and color variation. Also, their viability retention was estimated using the trypan blue method and their chlorophyll and carotenoids contents were measured by absorbance. All these methods gave us a better view on eventual damages due to processing conditions.

Results: ESD showed maximum preservation of pigments (both chlorophyll and carotenoids) in comparison to SD and FD. Also, both species were compatible with ESD by achieving good dehydration and colour values closed to initial color. Viability for all micro-algae was achieved maximum by FD but closely followed by ESD. Concerning particle size and span, there were almost similar for SD and ESD. Of course, particle structure for powders coming from FD was different due to the process. Morphology evaluated by scanning electron microscopy images showed no strong differences. All particles were dented or cracked due to rapid evaporation during the SD and ESD process.

Conclusion: For the first time, micro-algae were stabilized by an innovative process. Two chlorella species were successfully dried using ESD and this emerging process using electric voltage was found efficient to dry quickly micro-algae formulations.

Processing of Whole Okra Yield a Clean-Label Ingredient: Study of Thickening and Water Retention Properties

Doctor Marine Moussier^{1,2}, Marion Bruneau², Doctor Emma Donz², Professor Paul Menut¹

¹Paris-saclay Food And Bioproduct Engineering Research Unit, Inrae, Agroparistech, ²Ecole Supérieure des Agricultures

Aim:

Some additives currently used in food formulation, derived from multiple fractionation processes, lack natural alternatives. Okra (*Abelmoschus esculentus* (L.) Moench) is a widely consumed product across several continents, notably in Africa and Asia. Its pod contains over 80% water and approximately 11% pectin-like polysaccharides (mucilage), which impart interesting functional properties. These properties include thickening and water retention, rendering okra a promising ingredient for clean-label products. This study aimed to investigate these properties using whole okra powder obtained through minimal processing, with the objective of gaining a deeper understanding of the relationship between the properties of dried okra, rehydration performance, and the rheological characteristics of the resulting rehydrated products.

Method:

A whole okra powder was initially obtained through a simple process of drying and grinding, without the use of any solvents and without generating any waste or by-products. The powder obtained was rehydrated under different physico-chemical conditions (pH, temperature, ionic strength, etc.). Then, a combination of methods was used to investigate both the hydration properties of the powder, and the water retention properties, rheological characteristics (*i.e.* flow behavior) and structural properties (particle size analysis, microscopy) of the resulting matrice.

Results:

The whole okra powder disperses and rehydrates easily, releasing its mucilage without the need for heating. The results indicated that powder functionalization, *i.e.* mucilage release, is influenced by physico-chemical conditions. Particularly, an increase in ionic strength was observed to decrease mucilage release, while an optimal pH of around 7 was shown to enhance mucilage release. Once the powder dispersed in water and its mucilage solubilized, the resulting thick matrix showed both rheofluidifying and thixotropic behaviors. As expected, the texture level was directly linked with the quantity of powder and consequently the amount of mucilage. Lastly, analyses revealed the formation of a polymer network within the matrice.

Conclusion:

The whole okra powder is a clean-label ingredient for sustainable formulations requiring thickening power or water retention capacity. Further investigation is needed to understand the impact of physico-chemical conditions on the formation and properties of the mucilage network. Process optimization, including drying and grinding, offers a significant opportunity for improvement.

The shelf life of cold brew organic coffee

Eduardo Soares¹, Leticia Nogueira¹, Giovanni Bento¹, Carolina Machado¹, Eloiza Guerra¹, **Associate Professor Rodrigo Petrus**¹

¹Universidade de Sao Paulo Faculdade de Zootecnia e Engenharia de Alimentos

Aim:

The cold brew method consists of the cold infusion of roasted and ground coffee beans, resulting in a drink with low acidity and reduced bitterness. This study targeted the preparation, stabilization, characterization, and shelf life of a cold brew coffee drink prepared with organic beans subjected to three degrees of roasting: light, medium and dark.

Method:

The coffee beans were soaked in mineral water at 23 °C /24 h; the drink was then filtered, pasteurized at 90 °C /30 s, ultra-clean filled into 500 mL-transparent polyethylene terephthalate (PET) bottles and stored at 4 °C in the dark. Physicochemical, microbiological and sensory assays were carried out after 1, 35, 77, 119 and 150 days of storage. A team of 100 panelists evaluated the appearance, aroma, taste and the overall quality of the drink. Mean scores greater than 5 in a 9 point-hedonic scale and percentages of acceptance greater than 60% were set to estimate the sensory shelf life of the product.

Results:

The pH values varied between (5.0 and 5.3), (5.0 and 5.3), (5.2 and 5.7) respectively for the drinks prepared with light, medium and dark roasted grains. Regarding acidity, which was expressed as a percentage of chlorogenic acid, values between (0.011 and 0.039), (0.033 and 0.070), (0.027 and 0.061) were determined. The soluble solids content, in °Brix, ranged from (0.6 to 1.0), (1.4 to 1.9) and (1.4 to 1.9). The counts of aerobic mesophiles, psychrotrophs, and molds and yeasts were less than 1.1 logCFU/mL throughout the study.

Conclusion:

This finding demonstrated the microbiological stability of the drink for all degrees of roasting. The drink made with light roasted grains was rejected at the beginning of the study, in contrast to the medium and dark formulations, which achieved 150 days of shelf life.

Enhancing the Stability and Applicability of Olive Leaf Extract Through Double Emulsion Method

Demet Sönmezler¹, Nalan Yazıcıoğlu², Gulum Sumnu¹, **Prof. Serpil Sahin¹**

¹Department of Food Engineering, Middle East Technical University, ²Department of Nutrition and Dietetics, Gulhane Health Sciences Faculty, University of Health Sciences

Aim

Double emulsions are preferred for their superior ability to encapsulate and to protect sensitive ingredients, ensuring controlled release and enhanced stability. This research investigates the potential of this method to encapsulate olive leaf extract. Water-in-oil-in-water (W/O/W) double emulsions were formulated with varying concentrations of pea flour (15%, 20%, 25%) in the outer aqueous phase to evaluate their stability and effectiveness in containing the bioactive components. Pea flour was selected as a natural, plant-based emulsifier.

Method

Two-step emulsification method was used to prepare the W/O/W double emulsions. For the primary emulsion, inner water phase (W1) containing olive leaf extract was homogenized with the oil phase (O) containing the lipophilic emulsifiers. Ratio of inner water phase to oil phase was selected as 40% to 60%. In the second step, primary emulsion was further emulsified with the outer aqueous phase (W2) containing different concentrations of pea flour as the emulsifier. Ratio of primary emulsion to outer phase was again chosen as 40% to 60%. Characterization of the emulsions was done through analysis of their particle size, rheological properties, optical images, and encapsulation efficiency. For storage stability, emulsions were stored at refrigeration (4°C) and room temperature (20°C) for 2 months.

Results

Sauter mean diameter ($D_{3,2}$) of samples decreased while their apparent viscosity increased as pea flour concentration increased. Emulsions demonstrated high encapsulation efficiency, around 84%, effectively isolating the phenolic compounds from detrimental environmental conditions. The formulation with 25% pea flour exhibited superior stability among different concentrations with a stability of 91.22% at 4°C, over eight weeks. The better stability of emulsions prepared using 25% pea flour was attributed to the higher apparent viscosity and smaller particle size of the samples prepared with this concentration.

Conclusion

The findings highlight that double emulsions can serve as an effective delivery system for sensitive compounds, enhancing their stability and bioavailability in food applications. Our approach utilizes a natural plant-based emulsifier and thus promotes sustainable practices by utilizing agricultural by-products, contributing to advancements in food science and technology.

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Enzymatic hydrolysis of plant proteins from Norwegian-grown crops

Catrin Tyl¹, Thais Maria Rufino Barbosa, Emilie Gullberg Jørgensen, Tove Gulbrandsen Devold, Kenneth Aase Kristoffersen

¹Norwegian University of Life Sciences

Aim:

Plant-based milk analogues have become a popular alternative to milk in many countries. In Norway, they form the largest category of plant-based analogues to animal products. However, they are in many cases not produced from Norwegian raw materials. Moreover, their nutritional value is generally lower than that of cow's milk, especially in relation to protein and certain micronutrients (calcium and iodine). A challenge for increasing the protein content is the low solubility of many plant proteins. Therefore, this study assessed partial enzymatic hydrolysis of plant proteins obtained from Norwegian-grown crops and functional parameters related to applications in milk analogues.

Method:

The effect of two proteases was compared on fractions from air classification of faba beans, peas and oats. Endocut-01L or bromelain were used alone or in combination with alpha-amylase to treat 5 or 10% protein solutions for various times (2.5 – 180 min) at 55 °C and varying enzyme to substrate ratios. The degree of hydrolysis (DH) was assessed via the trinitrobenzenesulphonic acid method, protein solubility via the bicinchoninic acid assay. Protein profiles were evaluated via sodium dodecyl sulfate-polyacrylamide gel electrophoresis and size exclusion chromatography. Emulsion activity indices as well as emulsion stability were assessed as well.

Results:

At the lowest tested enzyme concentrations (0.5%), the DH did not change within the first 10 minutes. On the other hand, reaction times above 120 min only led to minor increases in DH, except for samples containing 10% protein. However, such samples were very viscous and as such difficult to handle. Due to the presence of starch in the samples, addition of alpha-amylase to decrease viscosity was assessed. However, it did not lead to increase in DH regardless of addition order, i.e., before, concomitantly or after the protease addition. In contrast to DH, protein solubility reached a plateau after 30 min and was not affected by starting protein concentrations (5 vs 10%).

Conclusion:

This study provides information on how hydrolysis via different proteases could increase protein solubility and how it affects interfacial properties. The hydrolysates may also be suitable for applications other than milk analogues if interfacial properties are improved.

The effect of anthocyanin structure on ternary anthocyanin-iron ion-pectin interactions and spectral properties

Valeria Weiss¹, Avi Shpigelman¹, Zoya Okun¹

¹Technion Israel Institute Of Technology

Aim:

Nowdays, consumer health awareness increased, and the trend toward healthier nutrition amplified the search for more natural food additives, such as natural colorants. Natural pigments such as anthocyanins are being explored to meet the demand for more natural food additives. However, they suffer from poor color stability during processing and/or storage compared to some synthetic counterparts; thus, new stabilization methods are being constantly explored. We aim to expand the knowledge regarding ternary pectin-metal ion-anthocyanin interactions, specifically the impact of anthocyanin structure on such interaction as a tool to stabilize anthocyanin color.

Method:

Iron-enriched pectin (PIr) was prepared by incubating apple pectin in an aqueous FeCl₃ solution to facilitate molecular "anchors" of iron on the pectin backbone. Pectin was characterized by ICP, and SEC-MALLS-RI. The effect of complexation (PIrA), and the effect of anthocyanin structure on coordination and color were studied using UV-Vis absorbance, color analysis, and Amicon Ultra Centrifugal Filters. The difference between blackcurrant anthocyanin extracts (multi-component) and their chemical standards (single-component) was studied as well.

Results:

At identical anthocyanin concentration, PIrA complexes resulted in blue color ($h_{ab}^* = 292.57 \pm 1.99^*$), compared to a light-pink aqueous solution ($h_{ab}^* = 322.00 \pm 19.71^*$). Bathochromic shift was observed as well. Lower K_d values were observed for delphinidins over cyanidins ($K_d(\text{D3G}) = 0.5 \pm 0.1^a [\mu\text{M}]$), $K_d(\text{C3G}) = 10.8 \pm 1.2^b [\mu\text{M}]$), containing three and two hydroxyl groups on their B-ring, respectively. The size of the attached sugar resulted in significantly lower K_d value ($K_d(\text{C3R}) = 27.1 \pm 2.8^c [\mu\text{M}]$), only for cyanidins. Furthermore, higher color stability was observed for the PIrA system ($K = 0.025 \pm 0.001^a [1/\text{day}]$), compared to the system where non-bound iron ions were added to pectin and anthocyanin ($K = 0.041 \pm 0.002^b [1/\text{day}]$). Empiric color degradation rates ($K [1/\text{day}]$) for PIrA were also structure dependent.

Conclusion:

The hydroxylation degree of the anthocyanin on the B ring and the glycoside type conjugated to the aglycon affects their interaction with PIr in a structure-dependent manner. On the other hand, when no iron ions are present on pectin, no interaction occur at pH=5. Due to PIrA complexation, sedimentation is prevented, blue color and color stabilization are observed, as opposed to apple pectin with added anthocyanins and Fe³⁺ ions (where Fe³⁺-anthocyanin complex formed without initially binding the iron to the pectin).

The effect of high sugar on the formation and stability of emulsion-based confectionery filling formulations

Miss Yanni Yang¹, Aris Lazidis², Isabel Celigueta Torres², Fotis Spyropoulos¹

¹University Of Birmingham, ²Nestlé UK Ltd

Aim

Emulsion microstructures are widely used to develop food products with unique sensorial characteristics. The use of emulsions to develop confectionery fillings provides more opportunities for new product development. There is an abundance of literature on emulsion formation, stability, and rheological behaviour of different food emulsions. However, emulsions in confectionery formulations are not widely studied. This could be attributed to their complex composition, the presence of high sugar levels and the requirement for a long shelf-life. Thus, there is an industrial interest in studying emulsion properties and stability under relevant sugar concentrations and storage conditions. Although investigation of physiochemical properties of emulsions in the presence of sugar has been reported (1,2), this is rarely carried out at sugar levels that apply to confectionery; ie > 40 w/w%. In addition, there is a growing research interest into the uses of plant-based ingredients in everyday foods responding to shifting consumer needs in terms of dietary requirements and sustainability. Such consumer trends raise the necessity to study the behaviour of plant-based emulsifiers in food emulsions. Moreover, research comparing animal-based emulsifiers with plant proteins within a high-sugar context is absent. This study aims to investigate the influence of different sugar concentrations, different types of emulsifiers on emulsion formation, stability and rheology.

Methods and results

The interfacial behaviour of different emulsifier types was evaluated at increasing sugar concentrations. The addition of sugar was found to impact the viscosity of the continuous phase, which further impacts emulsion formation. The role of the viscosity ratio on the droplet size and polydispersity was explored as a function of emulsifier type and sugar concentration. These parameters are found to further influence emulsion stability. The rheological behaviour of emulsions was also studied and interpreted regarding continuous phase viscosity, emulsion droplet size and emulsifier type.

Conclusion

Overall, this work fills in the knowledge gaps on the use of high concentration of sugar in emulsion production and its stability. It provides promising results for the successful development of emulsion-based confectionery fillings when considering their emulsion properties, stability and bulk rheological behaviours. In addition, plant-based emulsifiers are shown to have good potential for successful incorporation into confectionery formulations.

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Exploration and standardization of functional compounds from milk thistle cultivated in Korea

Seunguk Yu¹, Mr. Seongdo Lee¹, Mr. Hwanyun Lee², Dr. Dongyup Hahn^{1,2}

¹School of Food Science and Biotechnology, College of Agriculture and Life Sciences, Kyungpook National University, ²Department of Integrative Biology, Kyungpook National University

Aim

The compositions of silymarin, a group of flavonolignans found in milk thistle (*Silybum marianum*), may vary in extract depending on the varieties, cultivation methods, and environmental conditions. To establish a standardized methodology for evaluating milk thistle extracts as functional food materials, we explored the bioactive substances in milk thistle cultivated in Korea, where milk thistle was not cultivated in the past. We also aimed to analyze individual isomers, which is a known challenge in the analysis of milk thistle's composition.

Method

Milk thistle seeds (2.5 kg) obtained from the Rural Development Administration (Jeonju, Korea), were ground and extracted with 25L of methanol for 3 hours. The extraction was then partitioned using hexane, dichloromethane, and ethyl acetate, followed by separation using open column chromatography for non-polar fraction and Sephadex LH-20 resin. Compound isolation was achieved through preparative HPLC, while HPLC-PDA and LC-ESI-MS were employed for analytical purposes. NMR analysis was used for structural identification.

Results

By NMR and MS spectrum analysis, structures of flavonolignan isomers were confirmed, and it was observed that there are differences in the bioactivity of each of the compound depending on their conformation. Silybin and Silychristin were qualitatively confirmed in all solvent fractions, especially the most abundant in ethyl acetate fraction. Taxifolin and Epitaxifolin were isolated from ethyl acetate fraction. 2,3-Dihydrosilybin was qualitatively confirmed from hexane and dichloromethane fraction. We performed the detailed analysis through LC-MS/MS analysis and HPLC using chiral columns.

Conclusion

We confirmed the exact chemical structure and bioactivity of flavonolignans in milk thistle. This study is meaningful in establishing a methodology for measuring and standardizing the value of milk thistle extraction as a functional food materials.

Evaluating Protein Extraction Techniques from Lentils (*Lens Culinaris*): Comparing Ammonium Sulfate, Ultrasound-assisted and Conventional Extraction

M. Eng. Lisa Ziegltrum¹, Prof. Dr. Özlem Özmutlu Karslioglu¹

¹Weihenstephan-Triesdorf University of Applied Sciences

Institute of Food Technology at Weihenstephan-Triesdorf University of Applied Sciences is the coordinator of ProxIMed, a PRIMA project with 17 partners from 10 countries. The project aims to explore and implement products with alternative proteins in Mediterranean region. The proteins are extracted from sustainable, plant-based sources and novel foods using innovative and environmentally friendly processing technologies that have minimal impact on the nutrient content. Selected proteins are further developed into customized products.

Aim: The research aims to compare and evaluate the efficiency of environmentally friendly protein extraction methods from lentils. The focus is on the applications of conventional extraction, ammonium sulfate precipitation, and ultrasound-assisted extraction. Effectiveness is assessed by protein yield and potential applications in the food industry. Lentil proteins show great potential in milk alternatives and in other high-quality foods, owing to their valuable sensory and technofunctional properties.

Method: Yellow and red ground lentils are treated with three different extraction methods: (1) conventional extraction by alkaline solubilization and isoelectric precipitation, (2) ammonium sulfate precipitation, and (3) ultrasound-assisted extraction. Response Surface Methodology is used to optimize conventional extraction regarding pH, solvent-to-solid concentration (%w), extraction time, and extraction temperature. The other methods are adapted to the optimal parameters of conventional extraction. The extracted proteins are analyzed for yield, protein content, and technofunctional properties.

Results: Initial tests indicated that ultrasound-assisted extraction resulted in a higher protein yield ($65.5 \pm 1.7\%$) compared to conventional extraction. Optimization of extraction and processing parameters is expected to further increase the protein yield, as well as reduce chemical solvents used, and improve technofunctional properties. Other studies have proved that ultrasound is able to enhance the extraction from sources such as wheat germ. The limited research on lentils resulted only in one study that showed increased protein yield. This highlights a research gap in fully understanding the potential of ultrasound for protein extraction from lentils.

Conclusion: This study compares three methods for protein extraction from yellow and red lentils. A high potential for efficient and sustainable ultrasound-assisted protein extraction is emphasized. The resulting protein extract is tested for its technofunctional properties concerning its application in the food industry.

Characterization of Novel Musilage Extracts From Chia Seed, Quince Seed, Flaxseed, and Okra

Yaşar Özlem Alifakı¹, Beyza Ursavaş², Merve Silanur Yılmaz³, Özge Şakıyan Demirkol², Aslı İşçi Yakan², Kazım Sezer¹

¹National Food Reference Laboratory of Türkiye, ²Ankara University, Food Engineering Department,

³Bitlis Eren University, Kanık School of Applied Sciences

Aim: Musilages are plant based polysaccharides that are widely used for a range of industrial applications owing to their characteristics. They are used as health promoting, thickening, emulsifying, gelling, and stabilizing natural agents. The aim of this study is to determine the effects of microwave assisted extraction (MAE), ohmic assisted extraction (OAE), and ultrasound assisted extraction (UAE) of plant musilages from okra, quince seed, chia seed and flaxseed.

Method: Optimum extraction conditions were applied to each material and extraction type. Water was used as solvent. Nutritional composition (total protein, total ash, and dietary fiber) of mucilages was determined according to AOAC procedures (2010). The monosaccharides (glucose, xylose, arabinose, galactose and mannose) were analyzed using a Dionex ICS-6000 system (Thermo Scientific, USA) with pulsed amperometric detection (HPAEC-PAD). Fourier transform infrared spectroscopy (FTIR) using a Bruker/Vertex 70 Hyperion Series spectrometer equipped with a diamond-composite attenuated total reflectance (ATR) cell. The spectra were obtained in the range of 400–4000 cm^{-1} at a resolution of 4 cm^{-1} . The thermal characteristics of the mucilages were analyzed in an inert atmosphere (nitrogen gas) with a flow rate of 20 mL/min and heating from 25 to 220 °C at a heating rate of 10 °C/min. Scanning electron microscopy (SEM) analysis was performed using a Zeiss Evo 40 microscope (Germany). The SEM was operated at 15 kV with a magnification of 1000.

Results: It was found that monosaccharides (glucose, xylose, arabinose, galactose and mannose) contents of the extracts depend mostly on extraction and material type. Ohmic method had the highest monosaccharides contents for okra and flaxseed extraction, while ultrasound and microwave were the best for quince seed and chia seed, respectively. FTIR spectra were quite helpful for showing the presence of the main constituents of musilages. Besides, SEM and DSC characteristics of the musilages affected different extraction methods significantly.

Conclusion: Novel extraction methods like ohmic, microwave, and ultrasound were superior to conventional method for mucilage extraction with their higher nutritional and chemical compound contents.

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Effect of demineralization rate on caseins emulsifying and foaming properties in simple and complex matrix

Khadija Florence Dabo^{1,2,3}, Anne Laure FAMEAU³, Christine CHENE¹, Gaëlle GUILLET⁴, Romdhane KAROUI²

¹Adrianor, ²Artois University, ³Univ. Lille, CNRS, INRAE, Centrale Lille, UMR 8207-UMET, ⁴Inleit Ingredients

Aim:

Environmental and societal challenges are forcing industries to look for more sustainable ingredients. The dairy protein market is dominated by caseinates obtained by total demineralization, which have a bad reputation among consumers as ultra-processed products. However, they possess excellent functionalities when applied to food products.

The caseinates are obtained from total demineralization due to the breakage of the phospho-calcium bonds of casein micelles by milk acidification. One strategy to improve its environmental footprint is to reduce the level of demineralization. Five demineralization rates were studied from 0 to 100%. We determined the impact of decreasing demineralization rate on their foaming and emulsifying properties of proteins in aqueous solution, and finally on their use to produce whipping creams.

Methods:

A multiscale approach was employed. Emulsifying properties were studied by measuring the caseins content at the interface and by surface tension measurements. Foaming properties were determined by using neutron scattering, image analysis and tensiometry.

Whipping creams were produced with UHT at pilot scale from a mixture of animal and vegetable fats and caseins with their decreasing demineralization rates. The resulting creams were characterized in terms of droplets size, stability, interfacial and rheological properties. Finally, foams were prepared from these creams by whipping, and we evaluated their overrun, stability and firmness.

Results:

We observed that native casein exhibited the highest foam stability (29% of foam remaining after 30 minutes) in comparison to caseinate with no foam after 10 minutes.

The caseinates were identified as the most effective emulsifiers (smallest fat globules: $0,81 \pm 0,01 \mu\text{m}$ and lowest viscosity ~ 20 cP). Finally, we compared the overrun, stability and firmness of whipped creams. Casein demineralized at 20 % gave the best whipped creams with an overrun of 280 % and no exudate after 24 hours.

Conclusion:

Reduced demineralization rates were found to decrease the emulsifying properties of casein in both solution and whipping creams. However, they led to improve foaming properties, enhancing overrun and stability. The study suggests that partially demineralized caseins could offer good performances for the food industry comparable to that of caseinate, which has the largest market share and is constantly increasing in price.

Uncovering hydrolysis strategies for black soldier fly: thermal and acidic pretreatments

Sandra Borges¹, Tânia Ribas¹, Ana Rosa², André Almeida³, Manuela Pintado¹

¹Universidade Católica Portuguesa, CBQF - Centro de Biotecnologia e Química Fina - Escola Superior Biotecnologia, ²SEBOL – Comércio e Indústria do Sebo, S.A, ³I.T.S. – Indústria Transformadora de Subprodutos S.A

Aim: Edible insects have arisen as one of the most appealing alternative sources of proteins due to their nutritional value. Insect rearing has several benefits related to environmental sustainability, specifically, insects produce fewer greenhouse gases, need less space and water, and have a high feed conversion efficiency. Due to its numerous potential uses in feed, cosmetics and biodiesel production, the black soldier fly (BSF) is one of the most studied edible insect species. Thus, the objective of this study was to devise a methodology for generating black soldier fly (BSF) hydrolysates via enzymatic hydrolysis, with a specific focus on assessing the impact of various pretreatment techniques.

Method: This study reports the effect of thermal and acidic pretreatments coupled with the application of a commercial proteolytic enzyme (alcalase) on the liberation of protein/peptide, the resultant peptide profile and antioxidant properties of BSF hydrolysates. Pretreatments that were applied to the raw material as strategies to improve the hydrolysis performance, were (i) 2% (v/v) acetic acid (ii) 2% (v/v) propionic acid or (iii) high pressure and temperature (121 °C during 15 min). The BSF hydrolysates were then produced using 1.0% (E/S) alcalase for 4 h at 50 °C.

Results: The BSF hydrolysates using alcalase showed ca. 8000 µg/mL of protein, 40000 µmol/L of free amino groups, and an antioxidant activity via ABTS method of ca. 9000 µmol TE/L. Thermal pretreatment resulted in a greater improvement of enzymatic hydrolysis, evidencing an increase of ca. 30% in protein release, and 20% in antioxidant activity of BSF hydrolysates accompanied by an increase in peptides with molecular weight less than 10 kDa. However, acidic pretreatments did not show significant improvements in enzymatic hydrolysis.

Conclusion: The use of thermal pretreatment on edible insects followed by enzymatic hydrolysis showed that is a promising technology to generate BSF hydrolysates. These hydrolysates hold great potential as functional and sustainable ingredient for food/feed applications.

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Dairy powders: in situ characterization of particle surface features

Pr Claire GAIANI¹, Pr Serafeim Bakalis², Amine Filiali¹, Pr Mogens Andersen², Dr Kirsten Malmos³, Dr Jennifer Burgain¹

¹Université de Lorraine (LIBio), FRANCE, ²Department of Food Science, University of Copenhagen, DK-1958 Frederiksberg, Denmark, ³Arla Foods aamba Aarhus, Denmark

Aim: One part of the AMUSE project (funding from Innovation Fund Denmark) is related to *in situ* measurements of milk powders surface evolution. The overall aim of AMUSE project is to ensure growth of exports of dairy powders, through a validated platform able to predict shelf life under real supply chains. For this purpose, real industrial powders were characterized at the nanoscale level in order to add data and develop mathematical models from the surface to the particle and the powder bulk scales.

Method: Dynamic *in situ* Atomic Force Microscopy (AFM) was used to follow particle surface topography (i.e. rugosity), softness (i.e. young modulus measurements) during storage events (i.e. T°C, humidity). Results obtained by AFM were crossed with XPS analysis, surface fat extraction and classical physicochemical characterisation of milk powders. With this *in situ* technique it was possible to follow the same area at the particle surface under various temperatures ramps (from 10 to 50°C) and/or relative humidities ramps (from 20 and 75%). Two milk powders were analysed : whole milk and skim milk powders (WMP and SMP respectively).

Results: Particle surface evolution was followed for the two powders during storage events. Coupling AFM and XPS enable us to precisely identify lactose crystallisation event and/or surface fat evolution. It appears that crystal apparition was impacted by the nature of the powder (SMP or WMP), the temperature (under or above the glass transition temperature), the ramp kinetics (regular increase or increase by plateau). In comparison, crystallisation measurements with the dynamic vapor sorption (DVS) equipment could not be characterized so finely.

Conclusion: The AFM equipment equipped with a climatic chamber (T°C and RH control) and developed by the team was able to successfully mimic storage conditions (in the ranges: 10-50°C and 20-75RH%). This tool provides essential clues to better link particle surface evolution and functional properties. For the first time, *in situ* measurements were performed on milk powders and linked to storage events.

Effect of mild protein extraction methods, including MIPEF, on Duckweed and its functional properties

Phd Patricia Maag^{1,2}, Özlem Özmutlu¹, Cornelia Rauh²

¹University Of Applied Sciences Weihenstephan-Triesdorf & TU Berlin, ²TU Berlin

Research into duckweed has been intensified since it has found that duckweed has the potential to provide essential amino acids and valuable antioxidants, offering excellent potential as an innovative healthy protein source. As part of the *Smart Indoor Farming* research project ("Science and Art in Bavaria", STMWK), an interdisciplinary team of scientists at HSWT (University of Applied Sciences Weihenstephan-Triesdorf) is establishing an optimized mild protein extraction process using moderate-intensity pulsed electric fields (MIPEF) (supplied by industrial partner Vitave Tech).

Aim: The aim is to develop sustainable and mild extraction processes to reduce the amount of chemical solvents. MIPEF technology is used for cell disruption, focusing on protein release. The subsequent concentration of proteins will entail applying mild processing techniques such as ultrafiltration or diafiltration. In addition, the functional properties of duckweed protein concentrate (*L. minor* and *L. gibba*) will be investigated to define new food applications.

Methods: Utilizing a batch processing approach, MIPEF is applied to disintegrate duckweed plant cell membranes for protein release. This method is directly applied to pre-crushed duckweed. MIPEF-Parameters such as electrical fields (1-5 kV/cm), number of pulses, and pulse width are considered. Demonstrating the effect of MIPEF-assisted protein release and mass transfer, the conductivity index before and after treatment, fluorescence microscopy to visualize the protein release, and protein measurement are presented. The effect of MIPEF on antioxidant capacities and polyphenolic content is evaluated through several methodologies (DPPH, FRAP, Folin-Ciocalteu assays). Moreover, functional properties (solubility, foaming-, and emulsion capacity) are investigated to see the effect of MIPEF pre-treatment.

Results: In previous trials with duckweed plants, remarkably high protein contents of 35-40% (dry matter) were found. Within the scope of current research, the protein release of MIPEF-treated duckweed at 3-5 kV/cm was 10-15% higher compared to the untreated reference. The process led neither to a rise in temperature nor to a significant change in the pH value. Ultrafiltration processes enhanced protein yields compared to isoelectric point precipitation.

Conclusion: Mild extraction processes of duckweed proteins using MIPEF and ultrafiltration methods increase protein release and enable the preservation of sensitive nutrients as no heat and harsh chemicals are required.

Utilisation of natural surfactants in skimmed-milk double emulsions: Improving low-fat Cheddar Cheese functionality and Sensory.

Miss Millie Preece¹, Dr Lynn McIntyre¹, Dr Evi Paximada², Dr Helen Pittson¹, Dr Karim Farag¹
¹Harper Adams University, ²University of Leeds

Aim:

The research investigates natural alternatives for synthetic surfactants in double emulsion (DE) within low-fat (LF) Cheddar cheese (CC), with particular emphasis on replacing Polyglycerol polyricinoleate (PGPR) with sunflower lecithin in the primary emulsion. It aims to assess the effects on both DE formation and incorporation in LF CC, while also evaluating functionality and sensory attributes. This investigation addresses the challenges posed by the poor sensory and functional qualities of low-fat cheese, alongside growing consumer demand for healthier foods and a preference for clean label surfactants.

Method:

Primary emulsions were prepared by partially replacing PGPR with Sunflower lecithin at 2% (w/w) in 40 : 60 milk fat to distilled water, using ultrasonic homogenisation at 70% amplitude for 5 minutes. Skimmed milk double emulsions underwent method development using a Silverson High Shear mixer to create a DE suitable for incorporation into CC. Chosen DE parameters were 35:65 (W₁/O:W₂) at 6,000 rpm for 10 minutes. These DE were incorporated into 30 L low-fat CC batches, where the cheese milks were standardised to 1.05% wt. and 3.7% wt. for LF control/DE and full fat control respectively. After four weeks maturation at 12°C in vacuum pack bags, CC underwent nutritional analysis, texture analysis, and functionality tests, as well as sensory evaluation using the free-profiling method.

Results:

The results demonstrate that the partial replacement of PGPR with Sunflower Lecithin in the primary emulsion was successful at a ratio of 1.5: 0.5 and 1 : 1 producing D_{4,3} of 3.686 µm and 3.843 µm respectively and stable for utilisation in DE. These were then successfully incorporated into skimmed milk DE, producing droplet sizes ranging from 14.246 µm to 17.980 µm. These DE were suitable for the incorporation into low-fat CC. Sensory evaluation and functionality tests indicated promising outcomes, suggesting that the use of these DE can aid improvement of low-fat cheeses using fewer synthetic surfactants in the primary emulsion.

Conclusion:

In conclusion, this study suggests that PGPR can be partially replaced with sunflower lecithin in the primary emulsion, offering a natural alternative for stabilising DE in the production of low-fat Cheddar. The successful incorporation of DE into CC with favourable sensory and functionality attributes opens avenues for further developing CC, with enhanced nutritional profiles by using fortification of water-soluble vitamins through the DE system.

Extraction of Phenolics From Olive Mill Wastewater Using Novel Methods and Evaluation of Environmental Effect

Dr. Yasar Ozlem Alifaki¹, Dr. Merve Silanur Yilmaz², Mrs. Irem Sude Dogru³, **Dr Ozge Sakiyan**³, Dr. Asli Isci³

¹Republic of Türkiye Ministry of Agriculture and Forestry, National Food Reference Laboratory, ²Bitlis Eren University, Kanik School of Applied Sciences, ³Ankara University, Food Engineering Department

Aim: Olives have a high antioxidant capacity making them a potential source of bioactive compounds. However, only a small percentage of the total phenolic content (approximately 2%) of the olive fruit passes into the oil during the extraction, while the largest amount is unfortunately lost in wastes (olive mill wastewater and pomace) causing a huge environmental pollution. For eliminating this situation some extraction methods were suggested for olive mill wastewater treatment. In this study, microwave assisted extraction (MAE), ohmic assisted extraction (OAE), and ultrasound assisted extraction (UAE) of phenolic compounds from olive mill wastewater by using deep eutectic solvents (DES) were investigated. Besides, the environmental effect of different procedures were also evaluated.

Method: In the extraction process, DES: water ratio and waste water:solvent ratio were kept constant at 1:1 and 1:5, respectively. The effects of treatment type (microwave, ohmic, ultrasound or maceration), solvent type (choline-chloride: formic acid (C:F), choline-chloride: glycerol (C:G), choline-chloride: acetic acid (C:A)) and solvent molar ratio (1:2, 1:3, 1:4) on total phenolic content (TPC), pH values, total monomeric anthocyanin contents (TMAC), polymeric colour, colour density, polymeric colour ratio, and phenolic profile of extracts were examined. In addition, FTIR chromatograms and COD values of extracts which had highest TPC were determined.

Results: The extracts with highest TPC (approximately 14.500 mg GAE/g extract) were obtained during ohmic and microwave extraction. They provided 93.75 and 87.5% time-saving compared to maceration and resulted in higher oleuropein, 2,4 hydroxyphenyl ethanol, and hydroxytyrosol contents. The bands found in the FTIR spectrum were associated with oleuropein, 2,4 hydroxyphenyl ethanol and hydroxytyrosol, too. As a last remark, the COD of microwave extracts showed that the microwave procedure achieved a 90% dephenolization.

Conclusion: DES which is prepared with choline chloride and acetic acid can be suggested as a novel green solvent for the extraction of phenolic compounds from olive mill wastewater with higher TPC. Moreover its utilization in microwave extraction resulted in highest rate of dephenolization which makes them a promising solution for olive mill waste water's adverse environmental effect.

Identifying Marker Compound of Lignans in *Schisandra chinensis* seeds and Exploring Optimized Analysis Methods

SeongDo Lee¹, Hae Na Chi², Hwanyun LEE², Dongyup Hahn^{1,2}

¹School of Food Science and Biotechnology, College of Agriculture and Life Sciences, Kyungpook National University, ²Department of Integrative Biology, Kyungpook National University

Aim: The primary goal of this study is to isolate lignans, a notable physiologically active compound from *Schisandra chinensis* seeds, and to explore compounds with health promoting activities. The research further aims to develop and refine an optimized extraction method, enhancing both yield and purity of lignans, to support the development of health-functional food.

Method: Extraction of compounds was carried out using dichloromethane and MeOH, followed by a degreasing step employing hexanes and MeOH. And then subjected to purification through open column chromatography to isolate specific compounds. We utilized NMR and LC/MS for detailed chemical structure analysis. The physiological activities of the compounds were investigated using assays targeting nitric oxide (NO), inducible nitric oxide synthase (iNOS), and nuclear factor kappa B (NF- κ B).

Results: We isolated nine dibenzocyclooctadiene lignan compounds from *Schisandra chinensis* seeds and assessed their physiological effects, identifying those with the highest activity. We developed and standardized an HPLC-based extraction protocol to maximize the yield of these bioactive compounds. Modifications to the extraction solvent, temperature, and duration significantly enhanced the efficiency and reproducibility of the extraction process.

Conclusion: This study has developed and refined extraction and analysis methods specifically tailored for *Schisandra chinensis* seeds. The optimized methods ensure a higher yield and purity of dibenzocyclooctadiene lignans, which have been found in abundance within these seeds. These lignans are notable for their potent anti-inflammatory and immune-enhancing properties, underscoring their significant potential in the health-functional food sector. This study opens the way for the development of new health functional food products, providing that *Schisandra chinensis* seeds are good food ingredients.

Optimization of mixture composition and drying temperature of newly developed instant cocoa powder beverages

Miss Kristina Tušek², Associate professor Davor Valinger¹, Mrs Tea Sokač Cvetnić¹, Professor Jasenka Gajdoš Kljusurić¹, Assistant professor Tamara Jurina¹, **Associate Professor Maja Benković¹**

¹University Of Zagreb, Faculty Of Food Technology And Biotechnology, ²Health Centre Krapina-Zagorje County

Aim: The aim of this work was to analyze the influence of the proportion of honey, oats and cocoa, as well as the drying temperature, on the physical properties of the powder mixture for the preparation of functional instant beverage, serving as healthier alternative to products currently available on the market. Oat flour was utilized as a filler, while honey was employed as a natural sweetener.

Method: The effect of the honey and oats proportion (40%, 50%, and 60%), cocoa (5 grams, 6.25 grams, and 7.5 grams), and drying temperature (50°C, 60°C, and 70°C) on the physical properties (moisture content, particle size distribution, bulk density, flowability, water activity, dispersibility, wettability, and color) of the powder mixture was analyzed. Twenty-seven independent experiments were performed according to a full factorial design. Response surface methodology (RSM) was utilized to analyze the relationship between independent and dependent variables. Data were simultaneously fitted to a second-order polynomial equation using the Statistica 14.0 software package (TIBCO® Statistica, Palo Alto, USA).

Results: The obtained results showed that the proportion of honey and oats has a significant effect on the color of the powder mixtures. Furthermore, the results of the RSM modeling indicated that the drying temperature significantly affects the moisture content, bulk density, flowability, water activity, wettability, and particle size distribution. Based on the desirability profile derived from the RSM predicted values, the process conditions were optimized. The optimal conditions obtained using RSM for the physical properties of the powder mixture are as follows: proportion of honey and oats 55%, amount of cocoa 6.25 grams, and drying temperature 70°C.

Conclusion: Varying the ratio of honey and oats can alter the visual appearance of the beverages, potentially influencing consumer perception and preference. Furthermore, adjusting the drying temperature can lead to changes in these properties, allowing for optimization of the beverage preparation process. Optimal process conditions represent the combination that maximizes the desired physical properties of the powder mixture, leading to improved quality and potentially enhancing consumer satisfaction with the final product.

A mucic acid ester gallate as an anti-diabetes inhibitor isolated from Indian gooseberry fruits

Dr. Thao Quyen Cao¹, Dr. Dongyup Hahn¹, Ms. Hae Na Chi¹, Mr. Seongdo Lee¹, Mr. Seunguk Yu¹, Mr. Hwanyun Lee¹

¹Kyungpook National University

Aim: Protein tyrosine phosphatase 1B (PTP1B) plays a key role as a negative and positive regulator of insulin and leptin signal pathway; therefore, it plays a major role in type 2 diabetes mellitus which has become a major global health emergency. Indian gooseberry (*Phyllanthus emblica* L.) is a fruit widely consumed fresh and processed into fruit products in subtropical areas and has been used widely for thousands of years as a traditional medicine in Asian countries. The mucic acid methyl ester gallate, first reported from this fruit in 2023, was revealed to possess antioxidant effects. However, there has been no research on its PTP1B inhibition activity that has been investigated until now. Herein, the PTP1B inhibitory effect of a mucic acid methyl ester gallate isolated from the extract of Indian gooseberry fruits was examined *in vitro* and *in silico*.

Method: The extract was separated using dianion HP-20 and the isolated gallate was purified by preparative high-performance liquid chromatography. Its structure was elucidated based on extensive spectroscopic analysis as well as the comparison with those reported in the literature. PTP1B effect was evaluated using ursolic acid as a positive control. A kinetic assay was used for identifying the characteristics of PTP1B inhibition and molecular docking simulation was performed.

Results: The isolated gallate exhibited significant PTP1B inhibitory effects with the IC₅₀ value of 16.3 μM and acted as a mixed inhibitor against PTP1B enzyme. Docking simulations were successfully performed with the binding energies for the gallate to allosteric and catalytic sites of PTP1B were -6.50 and -6.19 kcal/mol, respectively. The mixed inhibition was proven through two hydrogen bonds and interaction with two residues at the allosteric site and eleven hydrogen bonds and interaction with ten residues at the catalytic site.

Conclusion: These results suggest that the isolated gallate may be a promising reagent for future studies on the development of new diabetes inhibitors, thereby contributing the evidence proving the potential of Indian gooseberry fruit and its gallate for the discovery of functional biomaterials.

Profiling Non-Volatile Dietary Biomarkers in Irish Origin Ruminant Forage

Sarah Ellen Kearney¹, Nigel Brunton², Dilip Rai¹

¹Teagasc Food Research Centre, Ashtown, ²School of Agriculture and Food Science, University College Dublin

Aim: To Profile Non-Volatile Dietary Biomarkers in Irish Origin Ruminant Forage

Metabolites present in meat from animals fed with specific diets could be used as dietary biomarkers for food traceability, authenticity, and to provide evidence for the role of food in human health well-being. Little is known about the phytochemical classes and composition of mixed-species swards commonly grown in Irish ruminant pastures. This study characterises and quantifies metabolites in common forages, which serves as a primary step towards establishing dietary biomarkers in meat from animals fed with these forages.

Method:

A total of 11 feed forage samples including monocultures, mixed species, and silage were assessed. Samples were extracted using 60% acetone with a solid-liquid ratio of 1:20 for approximately 20 hours at room temperature. Following centrifugation (10 mins at 5000 rpm), the supernatants were collected for free phytochemical analysis whilst the residual precipitates were subjected to alkali and acid hydrolysis to release cell-wall bound phenolics. Spectrophotometric based assays, namely total phenolic content (TPC), total flavonoid content (TFC), and total carotenoid content (TCC) were performed on all extracts generated. Additional UPLC-MS/MS quantification for individual polyphenols and HPLC-UV for carotenoids were performed on the extracts.

Results:

Between 2-3 folds higher total phenolic, flavonoid, and carotenoid concentrations were observed in the mixed species forages than the individual monoculture grasses. This was further supported by a higher content of flavonoids namely, genistein, kaempferol, and quercetin in mixed species swards than that of the monoculture grassland species. Similarly, β -carotene and lutein were in higher abundance in the mixed species swards than in the monoculture swards.

Conclusion:

The results clearly showed higher concentrations of carotenoids and polyphenols in grasses from a mixed-sward compared to individual grassland species. Mixed species pastures may therefore provide phytochemicals in higher abundance for grazing ruminants compared to monoculture sward pastures. Some of these phytochemicals dominant in forages could be potentially be traced in meat, which can serve as a dietary biomarker of origin for a multitude of purposes including food authenticity and traceability.

Black cumin essential oil as a potential active stabiliser of plant oils during heating

Assoc. Prof. Dominik Kmiecik¹, Katarzyna Kuraszyk¹, Aleksander Siger¹, Magdalena Rudzińska¹

¹Poznań University Of Life Sciences

Aim: The aim of this study was to evaluate the potential of black cumin essential oils to reduce degradation of rapeseed oil during heating.

Method: Rapeseed oil was heated with oil from black cumin (200 ppm, 500 ppm and 1000 ppm). The reference sample was rapeseed oil heated without additives and with the addition of the synthetic antioxidant TBHQ (200 ppm). The heating process was carried out at 170 °C ± 10 °C for 6 h, in a deep-frying model. Changes in the fatty acid profile, tocopherol and phytosterol content, polar compound content and triacylglycerol polymers were determined in unheated and heated samples. The nutritional quality indicators of lipids (PUFA/SFA, atherogenicity index, thrombogenicity index and hypocholesterolemic/hypercholesterolemic ratio) were also determined.

Results: The use of black cumin essential oil in a stability study of heated rapeseed oil contributed to a reduction in undesired transformations of oil. Compared to the synthetic antioxidant TBHQ, even the lowest addition of black cumin essential oil (200 ppm) showed similar or slightly higher protective properties. Higher additions of black cumin essential oil stabilized rapeseed oil significantly better compared to TBHQ. The best results were obtained with an addition of 1000 ppm. Oils with addition of black cumin essential oil showed lower loss of tocopherols and phytosterols and less change in fatty acid profile. Heated oils were also characterized by a lower increase in the polar fraction and the dimers of triacylglycerols.

Conclusion: The additives used led to an increase in the stability of the heated oil. In addition, the advantage of the essential oil used was its good solubility in the oil. Regardless of the size of the additive, the process was fast and the resulting oils were homogeneous, without precipitates or suspensions. The use of essential oils in frying oils may be one way to improve the stability of frying oils. However, one problem that may arise with their use may be their effect on the organoleptic properties of the oil and the fried product. Their intense sensory properties, taste and aroma, can affect the taste and smell of the fried food.

Impact of Processing on health-related compounds in selected Leafy Vegetables

Mr Robert Lugumira^{1,2}, Dr Geoffrey Ssepunya², Prof. Ann Van Loey¹

¹Ku Leuven, ²Kyambogo University

Aim: Leafy vegetables contain substantial amounts of phytochemicals of health importance, which are prone to degradation under specific processing conditions. These vegetables are usually cooked before consumption and dried in high production seasons to deal with their high perishability and scarcity during dry seasons. This study intends to evaluate the changes in health-related compounds (carotenoids, vitamin C, and phenolic compounds) resulting from cooking (steaming and boiling) and drying (sun, solar, and oven drying) of Leafy Vegetables.

Method: The most consumed Leafy Vegetables in Uganda, Red amaranth (*A. cruentus*), Green amaranth (*A. dubius*), African nightshade (*S. aethiopicum*), and Malakwang (*H. sabdariffa*) were selected and processed with the above techniques. Using HPLC, vitamin C and carotenoids were quantified, whereas spectrophotometric methods were used for the determination of total phenolic content (TPC), total flavonoid content (TFC) and total antioxidant activity (TAA) and comparisons were made with the raw samples to assess the processing effect.

Results: Cooking led to a reduction in total carotenoid content. Beta-carotene and lutein content, in particular, increased except for Malakwang, where lutein substantially reduced (>50% loss). Violaxanthin and neoxanthin were degraded by cooking, with higher impacts found in boiled samples. Vitamin C losses that ranged from 14-66% and 65-89% were noticed in steamed and boiled samples, respectively. Furthermore, boiling led to a reduction in total phenolic content (TPC), flavonoids (TFC), and antioxidant activity (TAA), whereas steaming caused no significant changes. On the other hand, drying decreased carotenoid contents, with a relatively higher decrease observed for violaxanthin. Drying led to no substantial changes in TPC, TFC, and TAA but caused noticeable losses in vitamin C, however, oven drying retained more vitamin C compared to solar and sun drying.

Conclusion: Steaming better retains health-related compounds in Leafy Vegetables compared to boiling. Oven drying is less detrimental to the health-beneficial phytochemicals in comparison to the other drying methods. Besides solar drying being able to protect the products from insect infestation and contamination, it has a similar effect on the phytochemicals as sun drying.

Food and nutrition security for Africa: harnessing underutilized crops and cellular agriculture for sustainable solutions.

Dr Belinda Meiring¹

¹Tshwane University Of Technology

Aim:

Achieving food and nutrition security remains a paramount challenge worldwide and with the 2030 deadline for the United Nations sustainable development goals approaching, there is a renewed urgency to find sustainable solutions. In Africa, the problem is exacerbated by population growth, climate change, and limited access to resources. Although the global expansion of high-yield, commodity crops increased food availability and reduced world hunger, there are still major challenges in addressing the “hidden hunger” and meeting dietary protein requirements. The challenge goes beyond providing nutritious food, having to meet the needs of the rapidly emerging middle class in developing countries in terms of consumer acceptability, affordability, availability and sustainability. This presentation explores innovative strategies centered around the utilization of underutilized crops and the principles of cellular agriculture to foster sustainable solutions.

Method:

Our research projects explored the potential of various nutrient-dense underutilized crops including Bambara groundnut, morula seedcake and Lowveld chestnut. Nutritional content, safety, quality and sensory properties were evaluated. We also ventured into cellular agriculture while participating in the XPRIZE “Feed the Next Billion” competition, developing a chicken breast alternative from laboratory-cultured chicken cells. Current research on alternative proteins focuses on increasing the protein content of mushroom mycelia cultured in liquid media.

Results:

Underutilized crops can improve food and nutrition insecurity in Africa, not only because they are nutrient-dense, but they are culturally relevant and have the potential to empower previously disadvantaged communities. The major challenges are inconsistent supply chains and developing modern processing technologies to create value-added foods. While cellular agriculture promises reduced environmental impact, improved animal welfare and enhanced food safety, there are major challenges with upscaling, reducing the production cost of cell-cultured meat and establishing legal frameworks. Growing mushroom mycelium in liquid media allows for efficient and sustainable production without the environmental impact associated with traditional agriculture. In addition, the nutritional value of the mycelia can be improved by optimizing the media composition.

Conclusion:

By harnessing indigenous underutilized crops and innovative technological approaches, we are working toward future food systems to provide safe, nutritious foods that are environmentally and socio-economically sustainable.

Linking molecular, microstructural and macroscopic changes to understand the cooking behavior of beans

An Nguyen¹, Patricia Namutebi¹, Ann Van Loey¹, Marc Hendrickx¹

¹Laboratory Of Food Technology, Center For Food And Microbial Technology, Ku Leuven

Aim:

Cooking is the key processing step in preparing bean seeds for human consumption. Because textural defects develop under adverse storage conditions, the influence of storage-induced aging on bean cooking behavior was considered as well. This study employs an integrated quantitative approach to comprehend structure-property relations in fresh and aged common beans during cooking.

Method:

The integrated methodological approach included quantitative changes at various length scales (molecular, microscopic, and macroscopic levels) during the cooking process of fresh and aged red kidney beans at 95°C. Molecular analysis included starch gelatinization, protein denaturation, and pectin solubilization. Microstructural changes were quantified using light microscopy coupled with image analysis, highlighting the evolution of cell expansion, cell separation, and starch swelling throughout cooking. At the macroscopic scale, texture (hardness) and volumetric swelling, identified as the two most prominent physical properties, were assessed. In view of a better understanding of the cooking behavior, correlations among the changes observed at the different length scales were examined.

Results:

The relationships identified among micro- and macroscopic properties effectively illustrate the profound impact of microstructure on texture degradation and swelling. The study confirms that bean softening strongly links to cell separation, pectin solubilization being the rate-limiting step in obtaining a palatable texture. Regarding volume changes, significant cell expansion during the initial stage of cooking largely contributes to the increase in cotyledon volume, while the role of cell separation becomes more pronounced in the later stages. Additionally, the seed coat, next to the cotyledon, emerges as a significant factor in the swelling of whole beans, which became less pronounced after aging. Molecular analysis suggests an insignificant contribution of starch gelatinization to bean softening, the complete gelatinization occurring in the early phase of cooking. Further swelling of gelatinized starch is suppressed through confinement by the cell wall, as indicated by microscopic findings.

Conclusion:

This study provides strong microscopic evidence supporting the direct role of the cell wall/ middle lamella network in microstructural changes during cooking of beans as affected by aging. Pectin solubilization drives cell separation, governing bean softening. These insights could be instrumental in structure engineering for tailored bean-based products.

Investigation of integral stereoselectivity of lipases on medium-chain triacylglycerol with chiral HPLC analytical method

Investigation Of Integral Stereoselectivity Of Lipases On Medium-chain Triacylglycerol With Chiral Hplc Analytical Method Jaehyeon Park¹

Investigation of integral stereoselectivity of lipases on medium-chain triacylglycerol with chiral HPLC analytical method Inwoo Park¹, Investigation of integral stereoselectivity of lipases on medium-chain triacylglycerol with chiral HPLC analytical method Jihoon Kim¹, Investigation of integral stereoselectivity of lipases on medium-chain triacylglycerol with chiral HPLC analytical method Juchan Lee¹, Investigation of integral stereoselectivity of lipases on medium-chain triacylglycerol with chiral HPLC analytical method Jaegwan Lee¹, Investigation of integral stereoselectivity of lipases on medium-chain triacylglycerol with chiral HPLC analytical method Juno Lee², Investigation of integral stereoselectivity of lipases on medium-chain triacylglycerol with chiral HPLC analytical method Pahn-Shick Chang^{1,2,3}

¹Department of Agricultural Biotechnology, Seoul National University, ²Research Institute of Agriculture and Life Sciences, and Center for Agricultural Microorganism and Enzyme, Seoul National University, ³Center for Food and Bioconvergence, Seoul National University

Aim: Establishment of analytical method for integral stereoselectivity of lipase on medium-chain triacylglycerol

The concept of integral stereoselectivity, which represents the selectivity of lipase against all acylglycerols during triacylglycerol (TAG) hydrolysis, was suggested. However, the analytical method to determine integral stereoselectivity has been established only for long-chain TAG. Because some lipases have limited activity on long-chain substrates, expanding the application of integral stereoselectivity to medium-chain TAG is necessary.

Method: Development of chiral HPLC system and interface-based kinetic modeling

We established the analytical method for the separation and quantification of tricaprylin, isomers of dicaprylin and monocaprylin, and caprylic acid. The caprylic acid and capryloylglycerols were separated using HPLC-UV/ELSD equipped with a single chiral stationary phase column, CHIRALPAK AY-3. Using four model lipases and a medium-chain specific lipase, the hydrolysis of tricaprylin was carried out. Through the kinetic model considering stepwise enzymatic hydrolysis and the characteristics of the oil/water interface, the kinetic constants indicating selectivity on each capryloylglycerol were estimated.

Results: Determination of integral stereoselectivity of lipases

Tricaprylin and its hydrolysates were successfully separated by the developed analytical method with more than 2.4 of all resolution factors. Time courses of tricaprylin hydrolysis catalyzed by lipases were obtained using the analytical system. The kinetic parameters were calculated by fitting the interface-based kinetic model with the experimental results. All four model lipases exhibited similar integral stereoselectivities to those observed for long-chain TAG, and the medium-chain specific lipase displayed *sn*-3 selectivity on tricaprylin.

Conclusion: Expansion of integral stereoselectivity for chain length-specific lipase

This study extended the substrate scope of integral stereoselectivity analysis to include medium-chain TAG, addressing the limitation posed by lipases with low activity on long-chain substrates. Our previous results and the developed methods in this study enable the investigation of lipase integral stereoselectivity regardless of fatty acid chain length, thereby contributing to a better understanding of the stereochemical characteristics of lipase.

A novel approach to green coconut water processing

Associate Professor Rodrigo Petrus¹, Alice Poça D'Água, Priscila Silva, Alessandra Oliveira

¹University of Sao Paulo

Aim:

Green coconut water (GCW) is a low-calorie drink rich in electrolytes such as potassium, magnesium and calcium, and is often used as an effective rehydration fluid. GCW is a relatively clear and colorless liquid. However, once the coconut is opened, the water begins to lose its nutrients and flavor, as well as changing color, partly due to the activity of endogenous enzymes, especially peroxidase (POD) and polyphenol oxidase (PPO). These enzymes catalyze changes in the flavor profile, as well as objectionable discoloration forming brown and pink pigments. This study primarily evaluated the combined effect of supercritical carbon dioxide (SC-CO₂) and mild temperatures on the PPO and POD deactivation of GCW.

Method:

A factorial design with 17 trials was performed to investigate the effect of temperature (in the range of 35 to 85 °C), pressure (75 to 370 bar) and holding time (13 to 47 min) on the enzymic deactivation, physicochemical parameters and color of GCW.

Results:

The percentages of reduction in PPO activity ranged from 3.7 to 100%, and POD ranged from 43.4 to 100%. The pH values of the freshly extracted and processed GCW were 5.09 and 4.90, and the soluble solids content were 5.5 and 5.4 °Brix, respectively. The holding time (t) had a significant effect ($p \leq 0.1$) on the total color variation. As for the reduction of PPO activity, the temperature (T) and the interaction between pressure (P) and t had a significant effect. None of variables (P, T or t) affected ($p > 0.1$) the POD reduction, pH and soluble solids variation.

Conclusion:

The findings herein indicated the viability of SC-CO₂ treatment assisted with mild temperatures to deactivate PPO and POD in GCW. Small variations in pH and soluble solids content were found between fresh and processed coconut water; however, they were not significant. Only t, in the range studied, played a significant effect in altering the GCW's color. There was also a significant and positive effect of temperature and the interaction between SCO₂ pressure and t in reducing the PPO activity.

Nutraceutical Potential of Yellow Kiwifruit from Huatusco-Veracruz

Jhusua David Reina Garcia¹, Nutraceutical Potential of Yellow Kiwifruit from Huatusco-Veracruz
Diana Guerra Ramirez¹, Nutraceutical Potential of Yellow Kiwifruit from Huatusco-Veracruz Juan
Guillermo Cruz Castillo¹, Nutraceutical Potential of Yellow Kiwifruit from Huatusco-Veracruz Gustavo
Almaguer Vargas¹, Nutraceutical Potential of Yellow Kiwifruit from Huatusco-Veracruz Alvaro
Castañeda Vildozola²

¹Universidad Autónoma Chapingo, ²Universidad Autónoma del Estado de México

Aim: The kiwifruit is perhaps the most nutritious known among berries, being rich in vitamin C, K, a good source of dietary fiber, and minerals such as potassium, calcium, phosphorus, and low in calories. However, currently, there are no studies describing the nutritional quality of yellow pulp kiwis in high-altitude tropical climates in Mexico. The objective of the following study was to determine the nutraceutical potential through a phytochemical analysis of *Actinidia chinensis* var. *chinensis* fruits from the municipality of Huatusco-Veracruz and compare them with imported fruits.

Method: The pulp (inner and outer pericarp) was lyophilized (1 g), mixed with 80 % methanol (1:10 w/v) and adjusted to pH 3 with 5 % HCl. Each sample was extracted by vortexing (1000 rpm, 3 min), sonication (15 min), incubator shaking (150 rpm, 37 °C, 30 min) and centrifuged (2500 rpm, 15 min). The supernatant was brought to a final volume of 10 mL. With these extracts, the antioxidant capacity (AC) and the total phenol (FeTC) and flavonoid (FITC) content were quantified. CA ($\mu\text{mol ET g}^{-1}$ dry weight) was assessed with ABTS [2,2'-azino-bis(3ethylbenzothiazolin)-6-sulfonic acid, A-1888] and FRAP (reducing antioxidant power of iron) assays. The calibration curves (CC) of these were determined with Trolox in a range of 3.84 to 46.1 mg mL⁻¹.

The FeTC (mg EAG g⁻¹ dry weight) was determined by the Folin-Ciocalteu method adapted to microplates. The CC of gallic acid was prepared in the range of 0.0013-0.0113 mg mL⁻¹. The FITC (mg EC g⁻¹ dry weight) was determined according to Kubola and Siriamornpun (2011), and a CC was prepared with catechin 0.00103-0.0516 mg EC mL⁻¹. The information was analyzed with Student's t-tests with the statistical program Infostat version 2020 ($P \leq 0.05$).

Results: The content of total phenols, total flavonoids, and antioxidant capacity of the fruit extracts from Veracruz were statistically higher than those of fruits imported from New Zealand.

Conclusions: the bioactive compounds of kiwifruit are an alternative for the Mexican diet since their consumption can reduce the risk of suffering diseases caused by oxidative stress due to their high antioxidant potential.

A new formulation tool for the optimization of biotics stability in dry mixtures

Dr Cecile Sampsonis¹, Francois Machuron², Etienne Tourte³, Dr Mathieu Clement-Ziza⁴

¹Phileo By Lesaffre, ²LIST, Lesaffre Institute of Science and Technology, ³Gnosis By Lesaffre, ⁴Digital & Data, Lesaffre Operations

Aim:

Water activity is a well-known and important factor for the long-term preservation of biotics activity included in dry mixtures. Mathematical models of sorption isotherms are available to understand how active substances interact with excipients, so that their stability in mixtures can be determined. However, the accuracy of these models is variable in some cases with over-smoothing specific areas of interest of sorption isotherms linked with biotics stability stages. Therefore, we investigated the use of non-parametric regressions to define a robust tool for the modeling of sorption isotherms and simulation of virtual dry mixtures containing biotics.

Method:

Sorption isotherms at 25°C of single dry components (biotics and 20 excipients commonly used in human and animal nutrition) were performed with a DVS Intrinsic plus. Sorption isotherms of single components were fitted with parametric models described in the literature and with a non-parametric specific approach (LOESS - Locally Weighted Scatterplot Smoothing). Simulations of sorption isotherms of mixtures were then performed according to Peleg and Norman (1992) concept with application of LOESS. Experimental binary mixtures were used to validate the simulation results.

Results:

Single component sorption isotherms were mainly successfully fitted with standard known models. However, the use of non-parametric regressions was essential for some biotics, such as a *Bacillus velezensis* probiotic or dry phages, to keep all the information from the curve and perform accurate virtual combinations afterwards. Thus, experimental sorption isotherms of 2 binary mixtures including those biotics were found to be fully included in the confidence interval (95%) of the simulated sorption isotherms of the corresponding mixtures virtually generated.

Conclusion:

Non-parametric regression as LOESS is well adapted to complex sorption profiles for which no standard model works and can be applied to accurately simulate sorption isotherms of mixtures. By knowing the critical water activity of a biotic, it is possible to propose formulations that maintain it in favorable conditions for a long-time preservation in complex mixtures. Developed as an online and user-friendly interface, this tool (patent pending) allows to address the growing challenges of precision formulation including biotics with sustainable and tailor-made formulatory approaches.

Effects of drying time and sugar content on the quality properties of beef jerky

Vitor Andre Silva Vidal¹, Rikke Harveland Ølberg¹, Ida-Johanne Jensen¹, Jørgen Lerfall¹

¹Norwegian University of Science and Technology

Aim:

Drying and salting processes are widely used in the food industry and are efficient processes that increase convenience and shelf life, improve flavor, and maintain the organoleptic properties of food products. Beef jerky is one of the oldest and frequently consumed meat product that are preserved by drying and salting. It has a unique flavor, is easily produced, and needs no refrigeration due to its low water activity. Furthermore, beef jerky has a high protein content, and can be made utilizing different techniques including several ingredients, and variations in drying processes. Therefore, the present study was set up to investigate the effects of sugar content and drying time on the sensory and physicochemical characteristics of differently produced beef jerky.

Method:

The effects of sugar content (0.5, 1.0, and 1.5%) and drying time (2, 4, and 6 hours) on moisture content, water activity, texture profile, and sensory characteristics were investigated. The texture profile was analyzed using a Texture Analyzer TA-XT2 (SMS Ltd., Surrey, England) and expressed as hardness, springiness, cohesiveness, and chewiness. The sensory characteristics were evaluated utilizing check-all-that-apply (CATA) questions utilizing the terms: smoked aroma, meat aroma, rancid aroma, red color, brown color, dry appearance, smoked meat taste, umami taste, salty taste, sweet taste, bitter taste, after taste, sticky texture, crispy texture, hardness, fibrosity, tender texture, and juicy texture.

Results:

The drying time and sugar content significantly influenced ($p < 0.05$) the moisture content and water activity. The texture parameters (hardness, springiness, cohesiveness, and chewiness) of different beef jerky treatments were significantly affected ($p < 0.05$) by the drying time used in the processing. Furthermore, the sugar content and drying time significantly affected ($p < 0.05$) the sensory characteristics of the different beef jerky investigated.

Conclusion:

According to the results obtained, the drying processing and sugar amount added in the beef jerky treatments impact the quality properties of the final product.

Cooking kinetics and mechanism of fresh and hard-to-cook common bean accessions of different market classes

Mr. Henry Tafiire^{1,2}, Boniface Odong¹, Irene Wainaina³, Robert Lugumira^{1,2}, Nguyen An¹, Patrick Ogwok², Tara Grauwet¹, Marc Hendrickx¹

¹KU Leuven, ²Kyambogo University, ³Jomo Kenyatta University of Agriculture and Technology

Aim: A large-scale study was conducted to obtain a more holistic understanding of the cooking behavior of beans, in terms of cooking kinetics and time to cook, as well as the mechanism of bean softening for different bean accessions.

Method: Fresh and aged soaked beans from twenty-four accessions were assessed for texture during cooking at 95 °C using the most informative objective compression texture analysis method. Texture evolution was modelled by the conventional fractional conversion model and modified three-parameter models to capture the lag-phase. Five accessions (slow to fast cooking) were selected to study the mechanisms of beans softening. A texture classification approach was used whereby the three main texture classes were assessed for texture evolution, starch gelatinization, protein denaturation and pectin solubilization during cooking.

Results: The initial texture varied across bean accessions and is significantly positively related to bean size. Significant but limited differences in initial texture on ageing were observed in majority of the bean accessions. Texture evolution, cooking kinetics, and time to cook varied across bean accessions and storage and all bean accessions developed the hard-to-cook defect. The softening rate constants for fresh beans were significantly higher and more variable than for aged beans. Fresh and aged beans of a given accession softened to a similar final hardness. Lag-phase was more prominent in aged beans than fresh beans. Incorporating lag improves performance of the conventional fractional conversion model. For all bean accessions, fresh or aged, starch gelatinization and protein denaturation were completed by the 10 to 30 minutes of cooking, respectively, far in advance of the texture levelling off to a final plateau value. The kinetics of pectin solubilization strongly align with the texture decay (softening) kinetics.

Conclusion: The difference in time to cook for fresh beans across accessions is mainly determined by the difference in initial hardness and cooking rate constant, while for aged beans changes in lag time and rate constant determine changes in time to cook. Softening during cooking of beans is mainly and directly influenced by pectin solubilization with protein denaturation having less influence and starch gelatinization having little to no influence.

First characterization of grapes cultivated in Guanajuato State, Mexico: harvests 2022-2024

María Elena Sosa-Morales¹, Nill Campos-González¹, Gerardo Fernández-Villanueva¹, María Susana Avila-García², Marco Bianchetti², Roberto Rojas-Laguna³, Stefano Toffanin⁴

¹Universidad De Guanajuato, ²Universidad de Guanajuato, ³Universidad de Guanajuato, ⁴Institute of Nanostructures Materials

Aim:

The state of Guanajuato in Mexico has an incipient wine industry. There are over 25 young vineyards in Guanajuato devoted to the cultivation of several grape varieties, most of them from France. The quality of the grapes, such as sugar content, acidity, and color, will determine the characteristics of the wines to be produced. Besides these physicochemical characteristics, grapes are recognized by their high content of phenolic compounds (PCs), which are related with antioxidant properties and can prevent some degenerative diseases in humans. The objective of this work was to follow the characteristics of grapes harvested in Guanajuato vineyards during three years, and to assure a good quality of the future wines produced from them.

Method:

Grapes (*Vitis vinifera* L.) were harvested from vineyards established in 2017-2018, located at municipalities of Guanajuato (around 1900-2300 m.o.s.l.) in the month of September of the years 2022 & 2023. They were transported in chilled plastic containers to the lab. Peel color with colorimeter, total soluble solids (TSS) with digital refractometer, pH with digital pHmeter, total acidity (TA) by titration and PCs with Folin-Ciocalteu reagent, were determined. Samples for year 2024 will be also analyzed.

Results: Grapes exhibited dark blue colours, with low values of lightness (L^* parameter), and negative values for b^* parameters. TSS were high, between 20.4 and 24.5°Bx, indicating a high amount of sugars to be converted in ethanol during the fermentation process for wine making. pH was in the range of 3.4 and 3.6, similar for all the assessed varieties. TA was the most affected variable by variety: Cabernet Sauvignon and Cabernet Franc grapes had 5.4-7 g tartaric acid/g depending on the year, while Syrah variety had the highest acidity ($p < 0.05$) with values between 8.5 and 9.2 g tartaric acid/g. Malbec and Merlot varieties showed similar acidity than Cabernet grapes ($p > 0.05$). PCs were in the range of 2.2 and 3.2 mg gallic acid/g fresh grape, without effect of the year.

Conclusion:

Grapes cultivated in Guanajuato State, Mexico, have good quality for producing red wines. It is important to follow them along years to seek effects due to vines adaptation.

Lipids act as a lubricant during the extrusion stage of pellet manufacturing, reducing agglomerate strength

Thomas Bastiaansen¹, Dr. Gerard Verge-Mèrida², Richard Benders³, Dr. Joshua Dijkman⁴, Dr. Menno Thomas⁵, Prof. Dr. Wouter Hendriks¹, Dr. Sonja de Vries¹, Dr. Guido Bosch¹

¹Animal Nutrition Group, Wageningen University & Research, ²Animal Nutrition Group, Institute of Agrifood Research and Technology, ³Physical Chemistry and Soft Matter Group, Wageningen University & Research, ⁴Van der Waals-Zeeman Institute, Institute of Physics, University of Amsterdam, ⁵Zetadec, ⁶Ruminant Production, Institute of Agrifood Research and Technology

Aim: The extrusion of materials, during densification processes such as pellet manufacturing, is a key step in converting bulky food or feed materials into dense agglomerates. The extrusion of materials containing high levels of fat, however, often yields physically weak agglomerates, but the underlying mechanism remains poorly explored. We investigated the effect of including various types of lipids, either added separately or as a part of the material, on the extrusion of feed pellets.

Method: Mixtures were formulated, containing, maize, soybean meal, and sunflower meal with increasing fat levels (0, 20, 40, 60 g kg⁻¹) by separately adding soya oil or poultry fat, or, to include lipids that reside within particles by adding sunflower expeller. Mixtures were conditioned and then extruded through a ring-die compactor. Energy consumption by the compactor was logged, and agglomerate strength was evaluated using compression tests. Data were analysed through linear regression analysis, with fat concentration and type as independent parameters. Fat distribution on the agglomerate surface was investigated using fluorescence lifetime imaging and fat dynamic viscosity was evaluated using dynamic mechanical thermal analysis.

Results: Fat coverage of the pellet surface appeared to be higher for agglomerates containing soya oil or poultry fat. Increasing fat concentration by including poultry fat reduced net energy consumption by the compactor (-0.033 kWh t⁻¹ per g kg⁻¹ of fat; $P = 0.007$) and a similar trend was observed for soya oil (-0.02 kWh t⁻¹ per g kg⁻¹ of fat; $P = 0.056$). Increasing fat content decreased agglomerate strength when including soya oil (-0.099 kN m⁻¹ per g kg⁻¹ of fat; $P = 0.047$) and poultry fat (-0.010 kN m⁻¹ per g kg⁻¹ of fat; $P = 0.021$). Increasing fat content by including sunflower expeller did not affect agglomerate strength or energy consumption by the compactor.

Conclusion: Our results suggest that separately added lipids migrate to the exterior of the agglomerate during extrusion, thereby acting as a lubricant between material and the die wall, reducing energy costs and agglomerate strength. The inclusion of lipids located within material particles, could allow for the production of strong agglomerates with a high lipid content.

Color and oxidation level of olive oil oleogels with ethyl cellulose of different viscosity

Claudia Armijo¹, Leticia Montes¹, Ramón Moreira¹, **Daniel Franco**¹

¹Department of Chemical Engineering, Universidade de Santiago de Compostela

Aim:

In recent years, food industry has opted to reduce TFA and SFA of their products, due to their link with cardiovascular disease and type 2 diabetes risk. New strategies using structuring agents such as ethyl cellulose (EC) by direct method, have been proposed. However, this method needs high temperature, which could affect the oleogel oxidative level, but this phenomenon is scarcely studied in the literature. Therefore, it was evaluated the impact of EC viscosity and concentration on color parameters and oxidative status of olive oil oleogels.

Method:

Commercially EC of different molecular weights, expressed by viscosity values (22, 46 and 100 cP) at different concentrations (8, 10, 12 and 15% w/w) were used for olive oil gelation. EC was dispersed on olive oil at 180°C for 1 h. Afterwards, the mixture was placed on foil-covered tray for cooling (20 min). Finally, oleogel was stored for 24 h, before analysis. The effect of both independent factors on color parameters (L*, a* and b*) and primary and secondary oxidation oxidative, measured by peroxide-value (PV), conjugated-dienes (CD) and p-anisidine-values (AV) on oleogel was assessed.

Results:

The findings showed that EC viscosity had the highest impact on L* and a*, although there was not a clear pattern. Concerning primary oxidation, there was a PV decrease for all elaborated oleogels as EC concentration and viscosity increased, but there was no reach statistical significance ($P > 0.05$). Indeed, CD values decreased significantly ($P < 0.05$) from 16.66 to 10.85 $\mu\text{mol/g}$ for oleogels formulated with 8 and 15% EC. The EC viscosity had the same effect as the EC concentration concerning the values of CD values. Both viscosity and EC concentration showed a protective effect against secondary oxidation measured by AV. For instance, AV decreased significantly ($P < 0.05$) from 58.04 to 27.89 as EC viscosity increased from 22 to 100cP. When EC concentration was evaluated, the AV diminished ($P < 0.05$) from 48.19 to 19.01 for increasing EC concentration from 8 to 15%.

Conclusion:

It can be concluded that EC concentration had a protective effect on the primary and secondary oxidation of olive oleogels in comparison to olive oil samples, submitted at similar temperatures.

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Influence of ethylcellulose viscosity and concentration on olive-oil oleogel: Effect on oil-binding capacity and texture

Claudia Armijo¹, Leticia Montes¹, Ramón Moreira¹, **Daniel Franco**¹

¹Department of Chemical Engineering, Universidade de Santiago de Compostela

Aim:

Within the food industry, the use of PUFA-rich oils to replace SFA and TFA using structuring techniques is being developed. Direct gelation with ethyl cellulose (EC) is widely studied. Still, few comprehensive studies focused on the concentration effect employed and the use of different EC with different viscosities, using olive oil. Here, it was evaluated the impact of viscosity and concentration of EC on the oil binding capacity (OBC) and textural properties of olive oleogels.

Method:

Commercially EC of different molecular weights, expressed by viscosity values (22, 46 and 100 cP) at different concentrations (8, 10, 12 and 15% w/w) were used for olive oil gelation. EC was dispersed on olive oil at 180°C for 1 h. Afterwards, the mixture was placed on a foil-covered tray for cooling (20 min). Finally, oleogel was stored for 24 h, before analysis. The effect of both independent factors on OBC and texture (TPA) on fresh oleogel was assessed.

Results:

EC concentration was the primary factor affecting OBC and textural parameters. In the case of OBC trait, additionally, both factors were consistent, raising the OBC as EC concentration and viscosity increased. OBC increased significantly ($P < 0.05$) from 79.47% to 99.57% for EC concentrations of 8% and 15%, respectively. Supporting this fact, a strong ($r = 0.875$; $P < 0.01$) correlation between EC concentration and OBC was observed. Oleogel hardness and gumminess increased significantly ($P < 0.05$) with EC concentration, meanwhile viscosity increase displayed an opposite behavior. Specifically, hardness and gumminess showed a wide range among tested oleogels. Indeed, hardness (3.97N to 59.47N) and gumminess (1.99N to 28.30N) values were obtained in oleogels formulated with 8% and 15% EC concentration, respectively. Textural properties have been related to organogelator concentration. In this study, we observed Pearson positive correlations ($P < 0.01$) between EC concentration and hardness ($r = 0.935$), gumminess ($r = 0.857$) and chewiness ($r = 0.820$) and negative with springiness ($r = -0.469$).

Conclusion:

OBC and textural parameters are crucial in oleogel applications for the food industry, particularly regarding replacing and incorporating fats in final products. Our findings confirmed that the use of commercial EC with different viscosity and concentrations allows a wide range of oleogels with tailored properties.

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Elasticity and oil binding capacity of structured olive oil with chitosan-vanillin dehydrated under different conditions

Mario Lama¹, David Rey, Leticia Montes¹, **Daniel Franco**¹, Amaya Franco-Uría¹, Ramón Moreira¹

¹Department of Chemical Engineering, Universidade de Santiago de Compostela

Aim:

Oleogels are of high interest as promising substitutes for trans fats in foods. To elaborate oleogel, most promising techniques are based on indirect method, which requires a formulation of a O/W emulsion, drying and homogenization steps. The aim is to study the influence of the drying conditions and oleogelator concentration on the oil binding capacity (OBC) and rheological properties of chitosan-vanillin based oleogels with olive oil.

Method:

O/W emulsions (50:50 w/w) with different chitosan content (0.7 and 0.8% (w/w) and vanillin/chitosan ratio (1.3) were air-dried (from 50 to 80°C) and freeze-dried, FD, (-26°C and 0.1 mbar). Oleogels viscoelasticity (frequency sweeps from 0.1 to 10 Hz at 0.1% of strain) was determined with plate-plate geometry (50 mm, gap 1.0 mm) at 25°C. Oleogels were centrifuged at 12,500×g for 25 min at 20°C for OBC evaluation.

Results:

High concentration of oleogelator increased the strength (high elastic modulus, G') in oleogels dried at 50°C and FD, but gels were weak (G' at 1 Hz < 52500 Pa). Oleogels dried at 60 and 70°C showed the opposite behavior with G' values increased hugely (< 80,900 Pa). This fact means that the structuration of chitosan-vanillin matrix was promoted. No differences were found in oleogels dried at 80°C regardless chitosan content due to a strong superficial hardening of emulsions during drying that avoided the proper structuration resulting in a dramatic weakening ($G' < 44,000$ Pa). OBC of air-dried oleogels were good (> 93.4%), obtaining the highest values (> 97.4 %) employing 60 and 70°C during drying. OBC increased with drying temperature from 50 to 70°C, but decreased at 80°C to values similar at 50°C. FD oleogels presented the lowest OBC (< 93.0%). Independently of both drying temperature and chitosan concentration, OBC values above 96% were only obtained in oleogels with $G' > 50,000$ Pa. A threshold strength of gel enables to bind the olive oil in the chitosan structure is necessary.

Conclusion:

The adequate selection of drying temperature of tested oleogels is necessary to obtain specific gel strength and OBC values. In fact, it was established a relationship between elastic character and OBC of the chitosan structure.

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Effect of extrusion parameters on meat analogs produced from durum wheat and pea proteins

Vittoria Latrofa¹, Aleksei Kaleda², Davide De Angelis¹, Kadi Jakobson^{2,3}, Antonella Pasqualone¹, Carmine Summo¹

¹University of Bari “Aldo Moro”, Department of Soil, Plant and Food Science (DISSPA), ²TFTAK AS,

³Tallinn University of Technology

Aim:

Durum wheat cake is a milling by-product from semolina production. It contains bran, germ, and debranning fractions of durum wheat and is rich in essential fatty acids, which could be extracted. Then the remaining cake could be dry-fractionated to produce a protein-rich ingredient. In this study, we aimed to test this novel protein ingredient in food applications, specifically in plant-based extruded meat analogs. For this purpose, we blended the ingredient with pea protein isolate (25:75) to get overall protein content of 66%. Because in addition to the techno-functional properties of the raw material the resulting fibrous texture of meat analogs is highly dependent on the extrusion parameters, we also investigated the influence of pH, moisture, and screw speed.

Method:

The conditions for low-moisture extrusion were chosen according to full factorial experimental design and were 28–32% for moisture, 400–600 rpm for screw speed, and pH was adjusted from 6.9 to 7.5 with potassium hydroxide. The properties of extrudates were compared using texture profile analysis (TPA) and descriptive sensory analysis; the water and oil holding capacities were measured also.

Results:

The pH, moisture, and screw speed had a major influence on the physical properties of the extrudates. The water holding capacity increased with higher screw speed and pH. TPA hardness decreased at lower moisture content and higher screw speed due to increased puffing of the extrudate. Sensory moistness was highly correlated with water holding capacity ($r = 0.87$), and fibrousness was the highest at the highest screw speed and moisture content regardless of pH.

Conclusion:

The novel protein-rich ingredient produced by defatting and dry-fractionation of durum wheat cake was successfully applied in extrusion of meat analogs. The extrusion parameters had a significant influence on the resulting texture and, thus, could be optimized to produce a variety of textures.

Tailoring the rheological properties of fat-free fermented concentrated milk products using high-power ultrasound.

Romario Mendes¹, M.Sc. Nico Piskors, Prof. Dr.-Ing. habil. Jörg Hinrichs

¹Institute of Food Science and Biotechnology, Dep. Soft Matter Science and Dairy Technology, University of Hohenheim

Abstract

Aim: Fermented milk products, also referred to as milk protein-based microgel dispersions, with high protein (8 – 12%) and low-fat contents, such as skyr, fresh cheeses, and concentrated yogurts, are becoming increasingly popular. However, these products are often not consumed by themselves because of their very dry and firm textures. Strategies currently available to improve the textural properties of these products often have disadvantages. For example, mixing with yogurt lowers the protein content, and mechanical treatments may even lead to higher viscosities. Therefore, this thesis aimed to employ low-frequency high-power ultrasound as a post-treatment to tailor the textural attributes of a fat-free fermented concentrated microgel dispersion, namely, fresh cheese.

Method: A pilot-scale test set-up was used to allow a continuous and cyclic post-treatment with low-frequency ultrasound (20 kHz) of two fresh cheeses with 9 and 11% (w/w) protein, respectively, at a fixed temperature, pressure, and volume flow (5 °C, 0.15 MPa, and 84 L/h) and energy inputs ranging from 0 to 140 MJ/m³. Apparent viscosity and yield stress were determined from oscillatory and rotational rheology measurements. Volume-based particle size distributions and volume fractions were obtained using static light scattering.

Results: With increasing energy input, the volume fraction and apparent viscosity decreased linearly, while the yield stress showed an exponential decrease. Further, the particle size of the fresh cheeses decreased, while the polydispersity of the product was only slightly altered. The lower apparent viscosities and yield stresses were attributed to lower volume fractions of the microgel particles. Furthermore, storage stability over two weeks was investigated.

Conclusion: The results thus suggest that the application of low-frequency high-power ultrasound is a promising approach to achieve a spoon- to drinkable high-protein fermented milk product with a creamy appearance.

Thermal Characterization of Selected Philippine Cacao (*Theobroma cacao* L.) and Tablea

Dr. Julius Andrew Nunez¹, Larrie Antoni Adaptar¹, Dr. Maria Carmen Tan², Dr. Jasmine Ting², Richard Anthony Galian², Dr. Joan Candice Ondevilla², Dr. Aldrin Bonto²

¹University Of The Philippines Manila, ²De La Salle University

Aim: Cacao (*Theobroma cacao* L.) is a high-value crop grown in tropical countries. In the Philippines, cacao is also used to produce a traditional product from cacao liquor called tablea. In tablea production, it is important to investigate the physicochemical properties to evaluate its nutritional attributes, and product quality. Hence, this study investigated the thermal properties of cacao and Philippine tablea from three different locations.

Method: The thermal properties of cacao and tablea samples were determined using differential scanning calorimetry (DSC), and thermogravimetric analysis (TGA). Other characterization techniques employed were proximate composition analysis, gas chromatography-mass spectrometry (GC-MS), and Fourier transform infrared spectroscopy (FTIR).

Results: The melting profile indicated that cocoa butter polymorph form V was the native structure of the cocoa butter in all samples, which is ideal for cacao-derived products. Additionally, repeated rapid heating and cooling of the cacao and tablea samples favored the formation of cocoa butter polymorph forms of lower stability. The TGA profile of cacao and tablea revealed the temperature ranges attributable to the degradation of polysaccharides, cocoa butter, and proteins. The DSC and TGA thermograms of cacao and tablea counterparts indicate similar phase transitions and thermal degradation events.

Conclusion: This study provided insights into the composition and thermal properties of cacao and Philippine tablea, which can be used as a baseline for quality and product development in the country.

Utilization of oleogels as a solid fat replacer in plant-based meat analogues

Youngseo Park¹, Suyong Lee¹

¹Department of Food Science & Biotechnology and Carbohydrate Bioproduct Research Center, Sejong University

Aim:

Excessive intake of saturated fats and trans fats is being recognized globally as a contributor to diseases including cardiovascular conditions. Therefore, a great deal of efforts has been made to reduce their contents in processed foods. Recently, oleogelation technology, capable of solidifying liquid vegetable oils while preserving their chemical properties, has garnered significant attention and this phenomenon extends to the domain of plant-based meat analogues. Thus, the oleogels prepared with three natural waxes (candelilla wax, carnauba wax, beeswax) were prepared and utilized as a solid fat replacer in meat analogues of which physicochemical properties were characterized.

Method:

Oleogels were prepared by blending natural waxes with liquid vegetable oils at a weight ratio of 1:9. Their textural/thermal properties were characterized. Also, when they were then applied to replace coconut oil in meat analogues, their effects on the physicochemical properties of the meat analogues were evaluated before and after cooking.

Results:

The hardness of oleogels formed by mixing three natural wax (candelilla wax, carnauba wax, beeswax) gelators with different vegetable oil varied significantly. The oleogels made with flaxseed oil exhibited the highest hardness, while those made with olive oil showed the lowest. Additionally, the oleogels produced with carnauba wax had the highest melting point, whereas those made with beeswax had the lowest melting point. When the oleogels were applied as a solid fat replacer, there seemed to be no differences in visual appearance. However, they affected the textural properties of the meat analogues. In addition, the cooking properties of the meat analogues were also dependent on the use of oleogels.

Conclusion:

In this study, it was observed that the rheological and thermal properties of oleogels varied depending on the types of natural waxes and vegetable oils used. When applied to meat analogues, the oleogels made from three different types of natural waxes provide a firmer texture without compromising quality characteristics. The findings of this study are expected to provide useful indicators for the development of low-saturated fat meat analogues, enhancing consumer preferences in plant-based meat analogues.

Effect of sucrose substitution on the physical and rheological properties of dairy custard dessert

dr Johnny Ciancetta¹, dr Marco Faieta¹, **Prof Paola Pittia**¹

¹University of Teramo, Department of Bioscience and Technology for Food Agriculture and Environment

Aim: Dairy desserts are sweet, formulated products whose nutritional value is under debate due to the presence of fats and sugars. While sucrose affects flavour and texture, it plays a main role on the microbial and physical stability of the products both under refrigerated and frozen state. To enhance the nutritional profile, alternative biomolecules and ingredients with different technological properties to be used in dairy dessert products without compromising their sensory profile, are under investigation (e.g. dietary fibers, hydrocolloids small saccharides). With the aim to enhance the knowledge on the effects of the use as sucrose-alternatives, this study aims to evaluate the effect of the partial substitution of sucrose in dairy custard dessert by different ingredients on physical and rheological properties just after preparation and after storage in frozen state.

Method: A reference custard (R), made of whole milk, egg yolk, starch and sucrose, was modified by partial (25, 50 and 75 %) substitution of sucrose and the use of maltodextrin (DE 8-10) -MD, inulin-IN and trehalose-TR. To obtain the custards a standard procedure including an heating step at 85 °C was applied. Modified custards were characterized for colour, thermal and rheological properties, particle size distribution as fresh and after 45 days in frozen state (-18°C).

Results: Moisture content and a_w of the modified custards was no significant different than R despite the different formulations. Significant decrease of the YI (Yellow Index) was observed for all re-formulated samples due to the microstructural and surface properties due to the alternative ingredients. Glass transition of maximally freeze-concentrated solutions (T'_g) increased being higher in MD and IN re-formulated custards with a concentration dependent effect. Particle size of MD- and IN-custards was higher than R, while the TR-custard was similar to the R one. All samples were characterized by shear-thinning behaviour with a significant increase in viscosity in MD samples. After frozen storage all samples showed a decrease of storage and loss moduli except IN-custard.

Conclusion: Results could provide insights on the effect of sucrose substitution with alternative bulk ingredients for the development of innovative dairy products and their quality and stability.

Assessing enzymatic protein hydrolysis in wheat flour and gluten model systems

Ms Fatemeh Sadeghian-motahar¹, Dr. Julia Rodriguez Garcia¹, Dr. Béatrice Kuschel², Dr. Paola Tosi¹, Dr. Michael Merz³, Dr. James Osborne², Dr. Afroditi Chatzifragkou¹

¹Department of Food and Nutritional Sciences, University of Reading, Whiteknights, ²Nestle Product Technology Centre Confectionery, Haxby Road, ³Nestle Fundamental Research Centre, Lausanne

Aim:

Achieving a light and crispy wafer with a uniform structure from a fluid batter requires flour with a weak gluten structure. Enzymatic hydrolysis is an effective method to prevent gluten development during wafer manufacturing process, potentially allowing for the use of different types of flours. This study explored the effects of protease treatment on the chemical structure of proteins in gluten and wheat flour.

Method:

The degree of protein hydrolysis (DH) across different protease concentrations (0.13-0.47 U/mL), time (15-120 minutes), and temperatures (30-40 °C) was measured in wheat flour and gluten samples. Sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE) was employed on both enzymatically treated gluten and wheat flour to visualize protein fragments. Further exploration of changes in protein secondary structures was achieved through Fourier Transform Infrared (FT-IR) analysis. The free SH (sulfhydryl) group content was measured to assess alterations in disulfide bonding.

Results:

The optimal protease activity for hydrolyzing wheat flour and gluten was identified at 35 °C, with gluten exhibiting a higher DH (43.4%) than wheat flour (11.4%) at the highest enzyme concentration tested (0.47 U/mL). SDS-PAGE analysis indicated the disappearance of bands linked to high and low molecular weight glutenin subunits, along with gliadins, and the appearance of new bands under 20 kDa. FT-IR showed a reduction in α -helix and coils, accompanied by an increase in β -sheet and β -turn structures confirming the degradation of proteins due to enzymatic activity. This observation was further evidenced by the rise in free SH groups, highlighting changes in protein tertiary structure.

Conclusion:

The study confirmed optimum conditions for wheat flour and gluten proteolysis and concomitant changes in protein secondary and tertiary structure. Future work will focus on examining the chemical and physical changes that protease confers in the wafer batter and on the assessment of its rheological properties.

Combinatorial solidification approaches for the production of pea protein based meat analogue model masses

Till Schumacher¹, Monika Gibis¹

¹University Of Hohenheim

Aim:

Industry, science, and society have been witness to a shift in the production and consumption of conventional meat products towards plant meat substitutes. However, there are allergenic challenges associated with the use of wheat and soy protein in these products. Pea, with its low allergenicity, wide availability, cost-effectiveness and good emulsifying and gelling properties, is emerging as a promising alternative protein source. The objective of this study was to investigate the gelation properties of two commercial pea protein isolates induced by different cross-linking mechanisms. Here, the effect of a combination of temperature, ionic strength, pH, and enzymatic treatment as cross-linking mechanisms was of special interest. The commercial pea protein isolates differed regarding protein solubility (high/low) and water/oil holding capacities (high/low).

Method:

Meat analogue model masses were prepared by mixing 15% pea protein isolate with water and emulsifying 10% rapeseed oil into the mixture with a blender. In addition, the cross-linking mechanisms were applied at that stage. A total of 22 sequences were prepared and analyzed regarding their maximum hardness with texture profile analysis using 75% penetration. The commercial pea protein isolates PISANE™ B9 (Cosucra®, Warcoing, Belgium) and Empro E86 HV (Emsland Group®, Emlichheim, Germany) were used for this purpose.

Results:

The two different pea protein isolates were chosen for their difference in protein solubility with PISANE™ B9 showing solubility of 9.2% and Empro E86 HV with a solubility of 40.7%. Different cross-linking mechanisms and their combination as well as different pea protein isolate products result in different hardnesses with significant differences ($p < 0.05$). Differences in maximum hardness for Empro E86 HV and PISANE™ B9 were found.

Conclusion:

The addition of different cross-linking mechanisms resulted in different maximum hardnesses. In addition, the choice of pea protein isolate results in differences regarding maximum hardness. A selection of a combination of crosslinking mechanisms based on the difference in solubility of pea protein isolates can be used for the development of vegan meat substitutes and to reduce the number of additives to achieve structure.

Impact of heating treatment on covalent and non-covalent interactions between protein and phenolic compound combinations

Chang Zhou¹, Imca Sampers¹, Katleen Raes¹

¹Ghent University

Aim: Protein-polyphenol interactions (PPI) are fundamental to various biochemical and physiological processes, such as enzyme inhibition and reduction of oxidative stress, which have significant implications for food and pharmaceutical products. This study aims to elucidate the effects of thermal treatment on the covalent and non-covalent interactions between casein and soy protein isolate—proteins with notable differences in molecular structure and solubility profiles—and phenolic compounds, namely gallic acid and quercetin. The goal is to understand how varying heating temperatures influence these molecular interactions and their impact on the structural properties of the resulting complexes.

Method: Protein solutions of casein and soy protein isolate and phenolic solutions of gallic acid and quercetin were prepared. These were subjected to both non-covalent and covalent complexation processes at 25°C and 100°C. Complexes were analyzed using UV-Vis spectroscopy to assess molecular interactions and structural changes, while intrinsic and synchronous fluorescence spectroscopy were employed to explore changes in the protein microenvironments. Additionally, fluorescence quenching methods were used to quantify the binding interactions and stability of these complexes under different thermal conditions.

Results: UV-Vis and fluorescence spectroscopy revealed that thermal treatment significantly alters the interaction dynamics within the protein-polyphenol complexes. At 25°C, non-covalent interactions dominated, with minimal structural alterations to the proteins. At 100°C, there was a notable shift towards covalent bonding, particularly evident in the casein-quercetin and soy protein-quercetin complexes. This shift led to significant changes in the absorption and fluorescence properties, indicating more stable and substantial modifications to the protein structures. Fluorescence quenching analysis supported these observations, with increased quenching constants at higher temperatures indicative of stronger static quenching interactions.

Conclusion: The study demonstrates that thermal treatments critically enhance the stability and modify the structural dynamics of protein-polyphenol complexes through increased covalent bonding. These findings are crucial for designing and optimizing food and pharmaceutical products that leverage the functional properties of these complexes. Controlled thermal processing thus offers a valuable tool for tailoring the functionality and stability of products containing these bioactive compounds.

Development of vegetable-derived scaffolds to improve lab meat organoleptic properties.

Hao Zhu¹, Célia Ferreira¹, Francisco Goycoolea¹, Alan-Javier Hernandez Alvarez¹, Amin Sadeghpour¹

¹School of Food Science and Nutrition, Faculty of Environment, University of Leeds

Aim:

With the overgrowing population set to reach 11.2 billion people in the next 80 years the reliance on conventional protein sources (meat, meat derivatives, fish) to meet the demand is becoming unattainable. Besides those sources create a huge environmental burden. The alternative protein products, namely meat analogues presently in the market do not meet consumers expectations, mainly in terms of mouthfeel, flavor and texture. The main defy of meat analogues is how to obtain the meat like texture in a sustainable and cost-effective way. This study aims to decellularize vegetables and shape them into a highly porous 3D fiber scaffolds that will function as cells carriers for the lab meat industry.

Method:

A number of vegetable side-streams were decellularized by two different methods: mild treatment and harsh treatment. In mild treatment, a low concentration SDS solution was used as a detergent to solubilize both the external and nuclear membranes of the vegetables. In harsh treatment, besides using a high concentration SDS solution, Triton X-100 and Sodium chlorite was used to decellularize vegetables, which remove lignin and produce holocellulose. After chemical treatment, both mild and harsh treatment samples were freeze by liquid nitrogen to destroy the cell membranes again using crystals created by low temperature and then freeze-dry. Dried, Rehydrated, Diced, Macerated and Pressed vegetables were subject to those decellularization steps and kept after freeze-dry in 50ml sealed containers for further analysis.

Results:

The scaffolds were characterized in what regards: a) microstructure by using Scanning Electron Microscopy (SEM) imaging and micro elemental analysis by using Energy dispersive spectrometer (EDS); b) decellularization efficiency and Total Carbon, Nitrogen and Hydrogen by combustion analyser; protein content by Dumas method; DNA content and total fibre content and c) preliminary physical/mechanical analysis measuring their viscoelastic behaviour using a rheometer and ζ -potential as well as interfacial tension measurements.

Conclusion:

We will utilize upcycled vegetable side-streams to construct the scaffolds to contribute for food losses reduction, promote circular economy and reduce costs of final product.

Innovative protein-based dairy cheeses by extrusion

Dr Mikkel Lorenzen¹, Adrian Tica², Frans W.J. van den Berg¹, Søren K. Lillevang³, Eric Windhab², Lilia Ahrné¹

¹University of Copenhagen, ²Swiss Federal Institute of Technology Zürich (ETH), ³Arla Innovation Centre, Arla Foods a.m.b.a

Aim: Extrusion technology has gained industrial interest to texturize plant proteins, while applications to dairy proteins to create cheese are still limited. Texturization of renneted casein emulsion gels by flat sheet die extrusion aligns the para-casein network, elongating the fat globules in an anisotropic structure formation in the shear-flow direction. The presence of milk fat globules embedded into an isotropic para-casein network may strengthen the overall casein gel network by acting as active fillers in the structure of the gel. This study aimed to investigate the impact of milk fat at four concentrations (0.1, 7, 11, 18, and 27% w/w) on the structural, rheological, and functional properties of texturized renneted casein-based extrudates.

Method: A co-rotating twin-screw extruder was used to texturize the casein gels and the extrusion process was monitored. Confocal laser scanning microscopy (CLSM) and X-ray microtomography, low-field nuclear magnetic resonance (LF-NMR), and small dynamic oscillatory measurements with temperature sweep were combined to characterize the anisotropic casein gel networks with different fat content at different length scales. The mechanical properties were assessed by a compression test.

Results: A higher fat content decreased the specific mechanical energy from 70.27 ± 4.55 to 61.42 ± 2.90 kJ kg⁻¹ due to the lower viscosity of higher-fat gels. LF-NMR measurements indicated that water molecules (T_{21} and T_{22}), on average, became more mobile with increasing fat content. Tightly bounded water (T_{21}) in texturized casein gels became more mobile compared to non-texturized casein gels. The small dynamic oscillatory measurements revealed that the internal strength of the casein network increased with increasing fat content, as indicated by the rise in storage modulus, for both non-texturized and texturized casein gels. The gel-sol transition temperature increased from 62.6 ± 0.7 to 66.0 ± 0.2 °C and from 64.0 ± 0.8 to 75.5 ± 0.7 °C with increasing fat content for non-texturized and texturized gels respectively. Microscopical observations revealed that the fat globule size and distribution influenced fiber formation.

Conclusion: Results showed the potential to create a variety of extruded cheese products by exploring the relationship between extruded cheese properties. The knowledge generated provided new insights to improve equipment design within the dairy industry and to produce novel cheese structures with customized behavior.

Vegan chocolate formulation using plant-based fat mixtures: understanding the effect of chemical composition on crystallization

Dr Elena Simone¹, Dr Cecilia Fiore¹, Tom Rutherford², Dr Stephanie Marty-Terrade³, Cynthia Marmet³, Francesca Giuffrida³

¹Politecnico Di Torino, ²Nestlé Product Technology Centre Confectionery, ³Nestlé Research

Aim: The EU chocolate market was worth around \$45 billion in 2022, and is constantly growing (<https://www.cbi.eu/market-information/cocoa/trade-statistics>). However, such increasing demand might lead to a significant negative environmental impact, since 1 kg of chocolate has a global warming potential of 2.91 - 4.15 kg CO₂ eq., primary energy demand between 30 - 41 MJ and water footprint ranging 31 - 63 L (Konstantas et al. 2018, Food Res. Int. 106, 1012-1025). The main contribution for this high carbon footprint comes from the raw materials, particularly milk powder, cocoa derivatives and sugar. For this reason, chocolate manufacturers have recently started to explore novel and sustainable solutions to replace or reduce the amount of these ingredients in chocolate. Cocoa butter and milk fat equivalents (CBE, MFE) are mixtures of triglycerides extracted from different sustainable plant-based sources (e.g., sunflower, shea) that resemble these two fats in their physical properties.

Method: In this work we applied synchrotron small and wide angle X-ray scattering, combined with polarized light microscopy and differential scanning calorimetry, to investigate the crystallization behaviour of selected commercial CBEs and MFEs, both on their own and mixed with cocoa butter. Cooling crystallization experiments were performed in the absence and with shear, and the number and type of polymorphs nucleated was studied.

Results: Experimental results were explained in light of the chemical composition of each fat mixtures, determined with chromatography. It was found that even small differences in triglycerides composition could dramatically affect the number, type and kinetics of crystallization of the polymorphs characterizing each sample. Additionally, significant differences in the crystallization behaviour were detected among samples with similar thermal behaviour and solid fat content. Shear was found to favour the nucleation or accelerate transformation into stable β polymorphs.

Conclusion: These results show the profound influence of triglyceride composition on the crystallization behaviour of complex fat mixtures. Hence, a detailed knowledge of chemical composition is necessary to understand at the molecular level the crystallization of fat mixtures, in order to efficiently replace CB and MF with their respective equivalents and rationally design novel vegan chocolate recipes that uses responsibly sourced, plant based fats.

METROFOOD.BE - Preparing the Belgian METROFOOD node to implement sustainable metrology services for food safety

Project Officer Charlotte De Bruyn¹, Subhalakshmi Sharma¹, Joris Van Loco¹

¹Sciensano

Aim:

METROFOOD.BE is the Belgian node of the METROFOOD Research Infrastructure (RI) that brings together high-level metrology services in food and nutrition. The aim of this project is to prepare the Belgian node, led by Sciensano, for the upcoming implementation phase of the METROFOOD-RI. This initiative aims to strengthen and integrate services related to food safety and metrology, ensuring a robust, state-of-the-art infrastructure that can address wide-ranging European and global needs in food analysis and safety.

Method:

Sciensano has been involved in transitioning METROFOOD from its design phase (PRO-METROFOOD) through preparation phase (METROFOOD-PP) and now towards the implementation phase (METROFOOD-EPI). Within METROFOOD.BE, the methodological focus is on strengthening existing service capabilities, improving user engagement through strategic testing and the development of use-cases, and enhancing methodologies to tackle essential food safety challenges. Sciensano will further enhance its services related to nanoparticles and food contact materials, emphasizing long-term sustainability and the development of a service provision framework that is excellence-driven, integrated, cutting-edge, and economically advantageous for its users. This will also involve streamlined integration into European research frameworks and ensuring compliance with open science and data principles. Additionally, efforts are being made to expand the Belgian node by attracting new Belgian research partners.

Results:

During its participation in earlier phases, Sciensano developed and refined methodologies for assessing food safety risks related to nanoparticles and food contact materials. These services have been recognized and utilized by various European entities, providing a foundation for further development of METROFOOD.BE. The proposed advancements will further strengthen their applicability and accessibility to a broader user base, including industry stakeholders and regulatory bodies. It will also offer more diversified and advanced metrological services within Belgium, thereby increasing its utility and reach within METROFOOD-RI.

Conclusion:

The development of METROFOOD.BE into METROFOOD-RI significantly contributes to the overarching goals of enhancing food safety infrastructure across Europe. By expanding the Belgian node, METROFOOD.BE will create a more robust infrastructure that supports advanced research, promotes safer food practices, and fosters innovation across the continent, solidifying Belgium's central role in the European food safety landscape.

Assessing Microbial Strain Variability and Kinetics to Ultrasound Treatment under Dynamic Temperature Conditions

Esther Okafor¹, **Dr Foteini Pavli**, Joerg HummerJohann², Vasilis Valdramidis^{1,3}

¹Department of Food Sciences and Nutrition, University Of Malta, ²Division of Food Microbial Systems, Agroscope, ³Department of Chemistry, National and Kapodistrian University of Athens

Aim

Ultrasound (US) is a promising non-thermal technology known for its antimicrobial efficacy. However, existing studies have not fully addressed the impact of strain variability and inactivation kinetics on US efficacy.

Method

Strain variability and inactivation kinetics were assessed by exposing four common food-associated microorganisms to US treatment under dynamic temperature conditions (26 kHz, 200 mL, 100% amplitude, 200 W, 65-71 W/cm²). Regarding strain variability, ten strains of *Listeria monocytogenes*, *Lactiplantibacillus plantarum*, *Saccharomyces cerevisiae* and *Escherichia coli* were exposed to US treatment. Samples were drawn before and after US treatment to determine microbial concentration. The same protocol was applied for the microbial inactivation kinetics of the selected resistant/sensitive strains. Thereafter, the log survival data was modelled under non-static conditions using the log-linear model and the Bigelow model as primary and secondary models, respectively, using a one-step approach.

Results

Strain variability assessments showed significant intra-species resistance ($p < 0.05$). *L. monocytogenes* strains L6 and NCTC 10357 were the most resistant and sensitive strains, respectively, having a difference of ~ 3 log CFU/mL. Regarding *L. plantarum* spp, FBR04 strain emerged as the most resistant, with a reduction of ~ 4.4 log CFU/mL, while *E. coli* FAM21845, FAM21805 and FAM21843 (~ 2 log CFU/mL) emerged as the most resistant strains. On the other hand, the most resistant strains of *S. cerevisiae* were CBS 1544, AD 1890 and 077.0001 (< 1 log CFU/mL) while AD 2913 was the most sensitive (> 5 log CFU/mL). The survival curves of most strains exhibited an initial phase of insignificant microbial inactivation followed by a relatively fast linear inactivation. The modelling of the survival data showed that the estimated $D_{T,UI}$ -value for *L. monocytogenes* strains (221.56 ± 61.22) was significantly different from other species ($p < 0.05$) while z_T values showed equal sensitivity to US treatment as temperature changes.

Conclusion

Strain and species variability play a vital role in the inactivation efficacy of ultrasound. The findings on strain variability resistance and inactivation kinetics of these microorganisms are essential for food safety and will pave the way for further research on microbial response to US stress, risk assessment and optimisation studies.

Antibacterial effect of Maillard reaction products produced by D-xylose and L-phenylalanine during normal cooking process

Ms. Hitomi Hirakawa, Mr Hiroshi Ono², Ms Junko Shinozaki², Mr Kento Koseki¹, Mr Shige Koseki¹

¹Graduate school of agriculture, Hokkaido University, ²Nisshin Seifun Group Inc.

Aim:

Maillard reaction products (MRPs), which are produced by heating reducing sugars and amino acids, are known for their various functional properties including antibacterial effects. Our recent study revealed that MRPs derived from D-xylose (Xyl) and L-phenylalanine (Phe) have strong antibacterial effects on certain spore-forming bacteria. As a novel application of the MRPs, we hypothesized that heating of D-Xyl and L-Phe together as cooking ingredients would produce antimicrobial substances during cooking and inhibit the growth of spore-forming bacteria remaining in the dish. The present study aimed to investigate the antimicrobial activity of the MRPs produced in food components during normal cooking process.

Method:

D-Xyl and L-Phe were mixed at 40 mM in a commercially available retort soup curry or retort beef bowl and heated at 95 °C for 1, 2, or 3 h. An aliquot of *Bacillus cereus* and *Clostridium perfringens* spores suspension (100 µL) were added to the heated soup curry and the beef bowl (900 µL). The inoculated heated foods were incubated at 25 °C for 48 h under aerobic and anaerobic condition for *B. cereus* and *C. perfringens*, respectively. Viable cell numbers in the samples were determined by plate counting after incubation of 0, 12, 24, 36, and 48 h.

Results:

The growth of both bacterial species were significantly inhibited in the soup curry and beef bowl heated with D-Xyl and L-Phe compared with the heated samples without those ingredients. The results indicated that the antibacterial MRPs were successfully produced during cooking process. The growth inhibition effect of the MRPs increased with extension of heating time. The difference in viable counts after 24 h incubation between the samples heated with and without D-Xyl and L-Phe was 1.5 log CFU/mL (*B. cereus* in beef bowl heated for 1 h) at the minimum and 6.2 log CFU/mL (*C. perfringens* in beef bowl heated for 3 h) at the maximum.

Conclusion:

The antibacterial MRPs can be produced when D-Xyl and L-Phe are heated in food during normal cooking process. The results suggest that adding D-Xyl and L-Phe as ingredients for cooking could realize successful control of spore-forming bacteria.

Prevalence and Characterization of *Enterococcus* spp. in Pristine Surface Water

Ms SzeChin Lim¹, Ayaka Nakamura¹, Takashi Kuda¹, Hajime Takahashi¹

¹Department of Food Science and Technology

Aim:

Enterococcus spp., indicators of water quality, are a growing concern because of their pathogenicity, which has led to many studies on contaminated water. In this study, we aimed to determine the prevalence and antimicrobial resistance of *Enterococcus* spp. in surface water, including coastal and river waters, with a minimal ecological footprint.

Method:

Fecal indicator bacteria (FIB), including *Enterococcus* spp. and *E. coli* in 60 surface water samples were analyzed using the membrane filtration method. A total of 92 presumptive *Enterococcus* spp. isolated using Slanetz and Bartley media were identified by 16S rRNA sequencing and tested for antimicrobial resistance using the Kirby–Bauer disk diffusion method. The prevalence patterns of *Enterococcus* spp. in coastal waters in relation to hydrological and physicochemical parameters were also analyzed.

Results:

The maximum counts of *Enterococcus* spp. and *E. coli* were 140 CFU/100 mL and 1200 CFU/100 mL, respectively, whereas the minimum count of both FIB was less than 1 CFU/100 mL in the surface water. A marginal difference in *Enterococcus* spp. count compared to *E. coli* count was found in both types of surface water. The weak relationship between the analyzed FIB ($r_s = 0.355$) was reinforced by the higher prevalence of *Enterococcus* spp. (63%) than *E. coli* (26%) in the coastal waters. The predominant species found in clean surface water included *E. entomosocium*, *E. hirae*, *E. mundtii*, *E. faecalis*, and *E. lactis* with slight variations in percentages. Surface water isolates showed resistance to linezolid, erythromycin, ciprofloxacin, tetracycline, vancomycin, penicillin, and norfloxacin. In coastal waters, physicochemical factors had no effect; however, a correlation between *Enterococcus* spp. and precipitation was observed. Heterogeneous and sporadic *Enterococcus* species were observed at the same sampling site throughout the sampling months. July had the highest number of antibiotic-resistant *Enterococcus* spp., particularly the isolates resistant to linezolid.

Conclusion:

The results indicated that *Enterococcus* spp. are more consistent than *E. coli* in pristine surface water. Diverse *Enterococcus* spp. with antibiotic resistance suggest the transmission of resistance, even at low bacterial counts. This highlights the suitability of *Enterococcus* spp. as water quality indicators for the safe harvesting of shellfish from coastal waters.

Enhanced Stress resistance of *Bifidobacterium breve* by induction of stress proteins at near-zero growth rates

Angela Rocio Ortiz Camargo¹, Oscar van Mastriht¹, Roger Bongers², Kaouther Ben-Amor², Jan Knol^{1,2}, Eddy Smid¹, Tjakko Abbe¹

¹Wageningen University and Research, ²Danone Research

Aim:

Bifidobacterium breve is a common habitant of the human gut and is used as probiotic in functional foods. *B. breve* has to cope with multiple stress conditions encountered during processing and passage through the human gut, including high temperature, low pH and exposure to oxygen. Additionally, during industrial processing and in the gut, *B. breve* could encounter nutrient limitation resulting in reduced growth rates that can trigger adaptive stress responses. For this reason, it is important to develop culture methods that elicit resistance to multiple stresses (robustness) encountered by the bacteria. In our previous study, using *B. breve* NRBB57 as a model, we determined the impact of near-zero growth rates in a lactose-limited retentostat on cellular parameters including culturability, viability, and metabolic pathway activation using a combined proteomics and metabolomics approach. The aim of the current study was to extend this analysis by determining the impact of imposed near-zero growth rates on *B. breve* NRBB57 stress defence proteomes and to identify correlations with stress resistance following exposure to (lethal) heat, low pH and hydrogen peroxide stress.

Methods:

B. breve NRBB57 was grown in lactose-limited chemostat cultures and in retentostat for 21 days, at growth rates ranging from 0.4 h⁻¹ to 0.00081 h⁻¹. Proteome analysis carried out at different growth rates were correlated to acid, hydrogen peroxide and heat stress survival capacity.

Results:

Comparative proteome analysis showed that retentostat-grown cells had significantly increased abundance of a variety of stress proteins involved in protein quality maintenance and DNA repair (DnaJ, Hsp90, FtsH, ClpB, ClpP1, ClpC, GroES, RuvB, RecA), as well as proteins involved in oxidative stress defence (peroxiredoxin, ferredoxin, thioredoxin peroxidase, glutaredoxin and thioredoxin reductase). Exposure to three different stress conditions, 45°C, pH 3, and 10 mM H₂O₂, showed highest stress resistance of retentostat cells sampled at week 2 and week 3 grown at 0.0018 h⁻¹ and 0.00081 h⁻¹.

Conclusion:

Our findings show that cultivation at near-zero growth rates induces higher abundance of stress defence proteins contributing to the robustness of *B. breve* NRBB57, thereby offering an approach that may support its production and functionality.

Exploring the Antimicrobial and Antioxidant Potential of Rapeseed Meal Extracts: A Promising Natural Remedy

Dr Sungkwon Park¹

¹Sejong University

Aim: Plants have long been integral to ancient medicinal traditions globally, offering a plethora of secondary metabolites utilized in medicine. Exploring natural ingredients such as rapeseed meal for their antioxidant activity presents a promising avenue for addressing contamination challenges in meat production industry. Rapeseed meal, a byproduct of rapeseed oil extraction, has garnered attention for its potential antimicrobial properties and radical scavenging ability. Our study aimed to investigate the antimicrobial potential and radical scavenging ability of rapeseed meal extracts, employing assays such as the DPPH assay, to evaluate its candidacy as a natural antimicrobial and antioxidant agent.

Method: Rapeseed meal extract was concentrated under reduced pressure and dried. Bacterial strains including *Bacillus subtilis*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Escherichia coli*, *Klebsiella pneumoniae* and fungal strains, including *Aspergillus niger*, *Candida albicans*, and *Curvularia lunata* were acquired from culture collections. The antimicrobial activity of rapeseed meal extracts was evaluated against the selected bacterial and fungal strains using disc diffusion method or broth microdilution assay. Zones of inhibition were measured after incubation, and minimum inhibitory concentrations (MICs) were determined using serial dilution methods. The radical scavenging activity of rapeseed meal extracts was assessed using the DPPH assay mixed with varying concentrations of rapeseed meal extracts at a specific wavelength (517 nm) after a suitable incubation period. Antioxidant activity was expressed as a percentage inhibition of DPPH radicals.

Results: Results revealed significant antimicrobial activity of rapeseed meal extracts against tested bacteria and fungi, with notable MIC values against *B.subtilis* (156.25 µg/ml), *S. aureus* (312.5 µg/m) and *E.coli* (156.25 µg/m). No inhibition was observed against *S. epidermidis* or *K. pneumoniae*. Phytochemical analysis indicated the presence of glycosides, flavonoids, and steroids in the extracts, suggesting their potential contribution to the observed antimicrobial effects. Additionally, rapeseed meal extracts exhibited substantial radical scavenging ability, as evidenced by their effectiveness in quenching DPPH radicals.

Conclusion: These findings underscore the potential of rapeseed meal as a natural source of antimicrobial and antioxidant agents, warranting further investigation into the isolation and characterization of its active constituents.

Identification of indicative taxa and optimization of cultivation to improve health in mung-bean sprout production

Dr. Luma Rossi Ribeiro¹, Dr. Karin Hassenberg¹, Tina Lütje², Dr. Lutz Hippe²

¹Leibniz Institute for Agricultural Engineering and Bioeconomy (ATB), ²MOL Katalysatortechnik GmbH

Aim: Mung beans (*Vigna radiata* L.) and their sprouts are widely consumed as fresh salad, vegetables or even as common foods. Sprout manufacturers face the challenge of meeting increased demand, on the other hand, climate changes and current political developments require a fundamental rethinking of how energy and water resources can be used efficiently. In addition, sprouts are classified as microbiologically critical, since the optimal growing conditions for sprouts (25 to 30 °C) are also favorable for the proliferation of pathogenic microorganisms. The goals of the project are, therefore, to minimize water and energy consumption while simultaneously increasing productivity and ensuring the microbiological safety of sprout production.

Method: For this purpose, a new generation of metal catalysts for water treatment is being developed by MOL (MOL Katalysatortechnik GmbH, Merseburg, Germany). In the first phase of the project, in collaboration with a sprout producing company, sprouts were evaluated at different stages of sprouting through microbiological, physiognomic, and molecular analyses (including PCR, gel electrophoresis, and sequencing). Additionally, MOL conducted an assessment of the water used in the sprouting process. From the microbiology side, mesophilic (total viable count), Enterobacteria, molds and yeast, *Bacillus cereus*, *Escherichia coli*, *Staphylococcus aureus* coagulase positive were evaluated.

Results: It was found ≤ 7 log/g and ≤ 5.3 log/g for mesophilic and molds and yeast, respectively. No *Bacillus cereus*, *Escherichia coli*, *S. aureus* coagulase positive were found. For the physiognomy, the sprout has gained approximately 2 to 3 cm (day 2 to day 7), with a final weight average of 0.6 g. Total protein content were also measured, and small variations were found in different days of sprouting, at day 6 protein content was 28.6 g/100 g (dw). For the water; pH, conductivity, mangan, chloride, Iron, sulfate and COD (chemical oxygen demand), was investigated.

Conclusion: The catalyst development is the next phase and aims to optimize the process of biocide-free industrial water treatment for mung bean sprout production. This process is intended to enhance sprout growth by improving nutrient availability in irrigation water, ensure a microbiologically safe product, and offer energy savings.

Rapid Detection of *Mycoplasma bovis*, *Staphylococcus aureus* and *Streptococcus agalactiae* in Cattle Bulk Tank Milk

Maria Liapi¹, Nikolas Markantonis¹, Christodoulos Papis², **Dr George Botsaris**¹

¹Cyprus University Of Technology, ²Cyprus Veterinary Services

Aim:

The aim of the study was to determine the prevalence of *Mycoplasma bovis*, *Staphylococcus aureus* and *Streptococcus agalactiae* in cattle farms in Cyprus and investigate the relations between the presence of the pathogens and the somatic cell counts (SCC).

Methods:

SCC were determined via flow cytometry, (Fossomatic™FC instrument, Foss, Hillerød, Denmark)

Bulk milk DNA extraction and Real Time PCR were performed using DNeasy Mastitis Mini Kit (Qiagen), Bactotype Mastitis HP3 PCR Kit (Qiagen), thermal cycler iQ5 (Bio-Rad). For *M. bovis* isolation and identification (DNA extraction, PCR) the following were used: *Mycoplasma* Experience solid and liquid Media (RH2 9BY Reigate, Surrey, UK), stereo microscope (Zeiss stemi SV 11) and QIAamp UCP Pathogen Mini Kit (Qiagen), PCR urvc gene 8, *Mycoplasma* spp. genus specific gene 9. For *M. bovis* identification and MLST typing with next generation sequencing the i-Seq 100 (Illumina) sequencer was used with Nextera DNA Flex Library Preparation (Illumina), and for the Bioinformatics analysis of the results obtained the Ccmetagen, (<https://cge.food.dtu.dk/services/CCMetagen/>) and pubMLST(<https://pubmlst.org/>)

Results:

One hundred and seventy-seven (177) bulk tank milk samples were analyzed with a commercially available real-time polymerase chain reaction kit and 11 (6.21%), 41 (23.16%), and 58 (32.77%) tested positive for *Mycoplasma bovis*, *Staphylococcus aureus* and *Streptococcus agalactiae*, respectively. Statistical analysis revealed a significant relationship between the presence of *S. aureus* and *S. agalactiae*. Enumeration of somatic cells was performed in the same samples by flow cytometry. The somatic cell counts were found higher in *S. aureus* and *S. agalactiae* positive samples. No association was found between *M. bovis* presence and somatic cells counts. Furthermore, low internal assay control Ct values were found to be related with high somatic cell counts. Finally, the likelihood of a *S. agalactiae* positive BTM sample to be found also positive for *S. aureus* was 8.9 times higher than the likelihood of a *S. agalactiae* negative BTM sample to be found also positive for *S. aureus*. No statistical analysis could be made for *M. bovis* (low number of positives).

The presence of *M. bovis* in Cyprus was unknown until this study. Therefore, samples from positive herds were cultured to isolate the pathogen and verify its presence in Cyprus. PCR identification and next generation sequencing of the isolates were additionally performed to confirm the presence of this pathogen and type it with MLST. According to the pubMLST database, the *M. bovis* ST type 29 was identified in this study, which has also been found in isolates from Israel.

Conclusion:

The real-time PCR in BTM is a useful, practical tool to detect mastitis pathogens in BTM. This study estimated a higher prevalence of *S. agalactiae* than *S. aureus* in Cypriot BTM samples from dairy cattle farms, and revealed the presence of *M. bovis* on the island, a neglected mastitis pathogen. Elevated SCC were confirmed to be related with real-time PCR detection of *S. aureus* and *S. agalactiae*, however they seem to have low usefulness for *M. bovis* infection. When real-time PCR is used as a diagnostic tool, low IAC Ct values can indicate a high SCC and even if a sample is negative for the pathogens tested, the presence of other mastitis pathogens should be investigated. Next generation sequencing is an extremely useful tool for one step identification or MLST typing of isolates.

The hidden power of plant leaves: Antibiofilm potential of vegetable-inspired biomimetic surfaces against foodborn pathogens

Mr. Fábio Carvalho^{1,2}, Rita Teixeira-Santos^{1,2}, Ana Azevedo^{1,2}, Maria Romeu^{1,2}, Cristina Amador³, Luciana Gomes^{1,2}, Kathryn Whitehead⁴, Jelmer Sjollema⁵, Mette Burmølle³, Filipe Mergulhão^{1,2}

¹LEPABE-Laboratory for Process Engineering, Environment, Biotechnology and Energy, Faculty of Engineering of University of Porto, ²ALiCE - Associate Laboratory in Chemical Engineering, Faculty of Engineering of University of Porto, ³Department of Microbiology, University of Copenhagen, ⁴Microbiology at Interfaces, Faculty of Science and Engineering, Manchester Metropolitan University, ⁵Department of Biomedical Engineering, University of Groningen, University Medical Center Groningen

Aim:

Biofilms are a potential source of product contamination in the food industry, reducing their shelf life and operational productivity. Therefore, there is an urgent need for comprehensive strategies for reducing bacterial adhesion and preventing biofilm formation. The superhydrophobic and self-cleaning properties of some leaves of water-repellent plants have been described to improve the antifouling performance of materials. This study aimed to produce and characterize the topographic and physicochemical properties of biomimetic surfaces of white cabbage (WC) and cauliflower (CF), and assess their antibiofilm performance against single- and dual-species biofilms of *Escherichia coli* and *Pseudomonas putida*.

Methods:

The biomimetic surfaces were produced by moulding using dental wax, and their thermodynamics were characterised by contact angle measurements, morphology by scanning electron microscopy, and roughness by optical profilometry. Biofilm formation assays were performed using 12-well microtiter plates for 24 h under static conditions, and the number of culturable and total cells, and biofilm architecture of both single- and dual-species biofilms of *E. coli* and *P. putida* were determined by colony-forming units, flow cytometry and confocal laser scanning microscopy, respectively.

Results:

The biomimetic surfaces presented cellular-shaped features, displaying successful replication of the micro-topographical features of the original leaves. They also presented higher roughness and hydrophobicity values compared to the flat control. These differences may have influenced the number of adhered biofilm cells, since the biomimetic surfaces were more efficient at reducing the numbers of bacteria on single-species biofilms (approximately 50% and 60% of reduction for *E. coli* and *P. putida*, respectively) compared to the control. Additionally, CF and WC surfaces reduced the biovolume, thickness, and surface coverage of single-species biofilms by 60 %, 45 %, and 60 %, respectively. In dual-species biofilms, both biomimetic surfaces showed a 60% reduction in the total biovolume and surface coverage of mixed biofilms. Moreover, *P. putida* outperformed *E. coli* growth in dual-species biofilms.

Conclusion:

These results highlight moulding as a reliable technique for reproducing the macroscopic topology of vegetable leaves and the potential of CF and WC biomimetic surfaces in reducing biofilm formation.

Behaviour of non- pathogenic *Escherichia coli*, Verocytotoxin E. coli O157:H7, O26:H11 serotypes during cheese process

Dr Elena Cosciani-cunico¹, Dr Elena Dalzini¹, Paola Monastero¹, Ehtesham_M Abdul¹, Daniela Merigo¹, Stefania Ducoli, Silvia Todeschi¹, Alessandro Norton¹, Marina_Nadia Losio¹

¹Istituto Zooprofilattico Sperimentale Della Lombardia E Dell'emilia Romagna "Bruno Ubertini"

Aim:

This study aimed to follow the growth and then non-thermal inactivation of non-pathogenic *Escherichia coli*, Verocytotoxin-producing (VTEC) O157:H7 and O26:H11 serotypes during the production of raw milk cheeses, by considering extrinsic and intrinsic variables that could affect the bacterial behaviour, implementing the predictive model's reliability and the dairy product risk assessment.

Method:

Nine challenge tests (experiments) were performed using 9 vats of 80 L of raw milk each. Milk was separately inoculated with *ca* 3 log CFU/mL-g of VTEC O157:H7 or VTEC O26:H11. Two test units were analyzed at each sampling time. VTEC concentration was determined on CHROMagar STEC medium, while non-pathogenic *Escherichia coli*, lactic acid bacteria (LAB) and physico-chemical variables by using ISO methods. During the cheese process, i) the log increase during the maturation (as the difference between the log concentration after maturation and the initial average log concentration), ii) the log inactivation after aging (as the difference between the log concentration after 14 weeks of aging and the average log concentration after maturation) were calculated.

Results:

In raw milk cheeses, the pH dropped to 5 ± 0.1 on the first day, the lactic acid bacteria concentration was 8.2 ± 1.5 log CFU/g and the lactic acid was around 10000 ppm. In 14 weeks of aging the *aw* decreased to 0.85 ± 0.02 . The non-pathogenic *E. coli* log increase ranged from 4.1 logs to 5.8 logs during the maturation, and a log inactivation from -1.5 logs to -2.5 logs was observed during the aging. The VTEC log increase was between 2.9 logs and 4.3 logs or between 3.67 logs and 4.75 logs, respectively for VTEC O157:H7 or O26:H11, with a log inactivation ranging between -2.4 logs and -4.2 logs or -2 logs and -4.5 logs, respectively for VTEC O157:H7 or O26:H11. The non-pathogenic *E. coli* behaviour showed a higher log increase during maturation and a lower log inactivation during 14 weeks of aging.

Conclusion:

The results suggest that non-pathogenic *E. coli* behaviour could represent the VTEC serotype during the raw milk cheese process, implementing the predictive model reliability and supporting the dairy products risk assessment concerning VTEC exposure.

Screening of the antimicrobial activity of Lactic Acid Bacteria against *Listeria monocytogenes* Scott A

Spiros Didos^{1,2}, Orfeas Saitis³, Zafeiro Aspidou³, Konstantina Tsotsouli¹, Konstantinos Koutsoumanis³, Anagnostis Argiriou^{1,2}

¹ Institute of Applied Biosciences, Centre for Research and Technology Hellas, ²Department of Food Science and Nutrition, University of the Aegean, ³Laboratory of Food Microbiology and Hygiene, Department of Food Science and Technology, School of Agriculture, Faculty of Agriculture, Forestry and Natural Environment, Aristotle University of Thessaloniki

Aim: The escalating threat of antimicrobial resistance necessitates innovative approaches to combat foodborne pathogens such as *Listeria monocytogenes*. Lactic acid bacteria (LAB) have emerged as promising candidates due to their inherent antagonistic properties. This study aimed to assess the antilisterial activity of LABs belonging to *Lactiplantibacillus plantarum*, *Lactiplantibacillus pentosus* and *Lactococcus lactis* species.

Method: Sixteen LAB isolates were screened through the in vitro agar spot assay to evaluate their antimicrobial activity against *L. monocytogenes*. The growth kinetics of co-cultures of nine LAB isolates with *L. monocytogenes* were assessed in liquid laboratory media in the presence and absence of glucose.

Results: *L. plantarum* species demonstrated the highest antilisterial activity according to the agar spot test followed by *L. pentosus* and *L. lactis*. On the other hand, *Lactococcus lactis* species indicated the highest suppression to the growth of *L. monocytogenes* followed by *L. plantarum* and *L. pentosus* when co-cultured with *L. monocytogenes* in BHI:MRS broth. Moreover, certain *L. lactis* spp. *lactis* strains exhibited enhanced inhibition effects when co-cultured in modified TSB (w/o glucose), compared to the other species that did not show significant inhibition against the pathogen.

Conclusion: This research underscores the potential of Lactic Acid Bacteria as natural antimicrobial agents for controlling *L. monocytogenes*. Harnessing the antimicrobial properties of LAB offers a sustainable approach to improving food safety and reducing the risk of foodborne illness associated with this pathogen, providing valuable insights and paving the way for the development of novel strategies to enhance food safety and public health.

Genomic and Phenotypic Analysis of Heat and Antimicrobial Resistance of *Escherichia coli* from Dairy Products

Dr. Christelle F. Iskandar¹, Dr. Reine Abi Khalil¹, Ms. Razan Zein Eddine¹, Dr. Antoine Abou Fayad¹, Pr. Mohammad G. Abiad¹

¹American University Of Beirut

Aim: Various microorganisms, especially *Escherichia coli*, have been associated with outbreaks caused by dairy products. *E.coli* developed heat resistance mechanisms to overcome the effect of pasteurization, in addition to the resistance to antibiotics. This study aimed to study the genomic and phenotypic characteristics of *E. coli* isolated from dairy products to explore their resistance to heat and antibiotics and their potential to be pathogenic and virulent.

Method: 63 dairy samples were collected from nonhygienic shops in Lebanon and tested for *E. coli*. The isolated cultures were subjected to *in vitro* heat resistance tests at 65°C for 30 minutes and 75 °C for 15 seconds. They were also tested against specific antimicrobial agents using standard antibiotic disks using the Disc Diffusion method. In addition, the isolates were subject to whole genome sequencing to search for genes responsible for their pathogenicity, virulence, heat, and antibiotic resistance.

Results: 19 samples were contaminated with *E. coli*, of which 13 were resistant to pasteurization treatments, and eight were resistant to antibiotics. Only two isolates from the heat-sensitive group were resistant to antibiotics. The isolates were shown to be pathogenic to humans, harboring a wide range of genes, among others, responsible for the isolate's capability to adhere to and invade the host. Hemolysins, toxins, and biofilm-encoding genes were also detected in many sequences. In addition, genes encoding stress-resistance proteins were found in isolates resistant to stress (heat and/or antibiotics) when absent in sensitive isolates' genomes.

Conclusion: The isolates detected in products sold in unhygienic shops were found to be human pathogens harboring a pool of genes, giving them the ability to be virulent and contributing to life-threatening food poisoning. Investigation should be done nationally to estimate the health risks to the general population.

Keywords: Dairy products, pathogenic *E. coli*, Heat Resistance, Antimicrobial Resistance, Genome Analysis

Risk-based evaluation of sampling plans for microbiological criteria to support risk management decisions

Leonardos Stathas¹, **Professor Kostas Koutsoumanis**¹

¹Aristotle University Of Thessaloniki

Aim:

Risk-based evaluation of sampling plans for microbiological criteria is crucial for supporting food safety risk management decisions. By adopting a risk-based approach, the food industry can strategically allocate resources and prioritize sampling efforts to areas of higher risk, ensuring more effective control measures and mitigation strategies. Such evaluations take into account factors like the severity of potential hazards, the likelihood of their occurrence, and the impact on public health, as well as the cost of microbiological criteria implementation. Through this process, decision-makers can optimize sampling plans to detect and address microbial contamination efficiently, ultimately safeguarding consumer health.

Method:

A 'sampling regime' module was incorporated in a risk assessment model (Stathas et al., 2024) so that the effectiveness of sampling tests in controlling foodborne risks could be evaluated. The sampling protocols tested were chosen based on EC No 2073/2005. The food-product combination tested was *Salmonella* spp. in chicken patties. The model was run in R programming language, using a Modular Process Risk Model (MPRM) methodology.

Results:

The results of the model show the relation between the acceptance criteria of the sampling plans (n , c , m) and the number of estimated salmonellosis cases in the European population. The baseline QMRA model for $n=0$ (no sampling) predicted a mean probability of illness of $1.19 \cdot 10^{-4}$ ($5.28 \cdot 10^{-5} - 3.57 \cdot 10^{-4}$ 95% C.I.), and a mean annual number of illnesses per 100,000 people of 2.13 (0.96 – 6.59 95% C.I.). The model assuming the implementation of the current sampling plan for *Salmonella* spp. in poultry (EC No 2073/2005, $n=5$, $c=0$) results in almost no impact (considering the uncertainty) on the cases of salmonellosis from the consumption of chicken patties compared to no sampling. Considering the cost of implementation of such a sampling plan, the later observation is of great importance for food safety management.

Conclusion:

Using the risk-based approach presented in this study, public health can be enhanced by applying strategic risk management to redistribute resources with the maximization of public health as the primary objective.

Magnetic nanoparticles based enrichment and colorimetric detection for Human Norovirus

DR Prabir Kumar Kulabhusan, Dr. Andrey Ipatov, Dr. Marta Prado

¹International Iberian Nanotechnology Laboratory (INL), Av. Mestre José Veiga s/n, 4715-330, Braga, Portugal., ²International Iberian Nanotechnology Laboratory (INL), Av. Mestre José Veiga s/n, 4715-330, Braga, Portugal., ³Department of Analytical Chemistry, Nutrition and Bromatology Faculty of Veterinary Science, Campus Terra. University of Santiago de Compostela, 27002 Lugo, Spain

Aim: Human Norovirus (NoV) is known as the primary cause of viral gastroenteritis and food-borne diseases, worldwide. Presently, there are no vaccination or antiviral treatments available for human NoV. Therefore, it is imperative to establish a timely and efficient method for the detection of NoV in order to mitigate its transmission within the food supply chain.

Methods:

To detect human NoV rapidly and sensitively in food matrices, it is necessary to concentrate and purify the viruses prior to detection. However, it has been a major challenge to concentrate human NoV in food matrices. In our study, we have employed procrine gastric mucin (PGM), aptamers and monoclonal antibodies as the bio-receptors for the pre-concentration of NoV. These receptors are conjugated to the magnetic nanoparticles (MNPs, ~100 nm) using various techniques such as streptavidin-biotin and EDC-NHS chemistry. The conjugation of the bio-receptor with the MNPs was confirmed using FT-IR and TEM. The conjugation procedure was optimized and studied for the enrichment of the NoV virus-like particles (VLPs) from the spiked samples. Furthermore, for the detection NoV, aptamer-conjugated MNPs were used to capture the NoV and the monoclonal antibody-conjugated gold nanoparticles (AuNPs) were utilized in a colorimetric assay. NoV was visualized by studying the peroxidase activity of the AuNPs with the one-step colorimetric analysis. Furthermore, a comparison was also done using antibody-conjugated MNPs and the aptamer-conjugated AuNPs as the detection probe.

Results: The comparison of three baroreceptors was studied for the capture of NoV. We found that monoclonal antibodies have higher capture efficiency as compared to the PGM and the tested aptamers. The characterization of receptor-conjugated MNPs were confirmed and protocol for NoV capture was optimized. Colorimetric assay demonstrated a good limit of detection (LOD) in the spiked samples.

Conclusion: Herein, different bioreceptors were studied for the capture of the NoV and a colorimetric method was developed for the visualization of NoV in spiked samples. The assay has to be studied in the real food matrices in the near future.

Is consumption of Brown Seaweed - *Alaria esculenta* Safe? Quality and Safety assessment during storage

Inthuja Manickam^{1,2}, Katharina Nøkling-Eide^{1,2}, Anita Nordeng Jakobsen¹, Ida-Johanne Jensen¹

¹NTNU Department Of Biotechnology & Food Science, ²SINTEFF Industry

Aim:

Seaweed utilization for human consumption is gaining attraction as plant-based food demand is at its peak. While selectively extracted components of macroalgae like carrageenan and alginate are used in the food industry, utilization of whole seaweed is less explored. One of the factors is the food safety aspect of seaweed preservation. Food processes like drying and freezing are used to ensure microbiology stability and a longer shelf life of macroalgae. Moving toward sustainable and energy-efficient processes in the food industry, the acidification of seaweed is explored taking into account its physicochemical, bioactive and sensorial properties at lower pH preservation. However, very little exploration is done on its microbiological stability during storage. Hence, the present study was focused on investigating microbial stability during storage at a lower pH.

Method:

Cultivated *Alaria esculenta* were blanched for 2 minutes at 80°C in acidified water. After the blanching, the pH was adjusted until a stable pH of 3.6 was achieved. The samples were then sieved to remove excess water before vacuum packing. Samples were stored at room temperature (RT) for a set duration. Samples were collected at a higher frequency in the beginning of storage period and after with lesser frequency based on microbial growth. Microbial analysis such as total aerobic microbial counts, total aerobic plate count of yeast and mould, Enterobacteriaceae, *E.Coli*, Lactic acid bacteria spoilage, *Listeria monocytogenes* and spores forming bacteria were investigated using methods recommended by the Nordic committee on food analysis. In addition, pH, water activity and moisture content were measured.

Results:

pH played a vital role in slowing and/or eradicating specific bacterial colonies in the samples while the vacuum pack limited the amount of air available for the proliferation of aerobic bacteria. Regardless, the trapped air and water presence within the folding of whole-blanched seaweed adds to the complexity of preserving for a longer duration at RT.

Conclusion:

Results indicate the possibility of preserving whole seaweed more sustainably. However, further research is needed to explore the sensorial and nutritional properties of seaweed stored after acid blanching to ensure the product is of acceptable quality and taste for consumers.

Modeling of Shiga toxin-producing *Escherichia coli* growth in minced pork meat

Doc. MVDr. Ph.D. Lenka Necidova¹, Doc. MVDr. Ph.D. Sarka Bursova¹, Mgr. Katerina Stojanova¹, RNDr. Ph.D. Danka Harustiakova², Ing. Ph.D. Klara Bartakova¹, Mgr. Ph.D. Alena Zouharova¹
¹University of Veterinary Sciences Brno, ²Masaryk University

Aim:

Shiga toxin-producing *Escherichia coli* (STEC) is a zoonotic gastrointestinal pathogen capable of causing moderate to severe diarrheal diseases with life-threatening complications, including haemolytic uraemic syndrome (HUS). In the last years, infections caused by STEC represent the fourth most common human zoonotic infectious disease in the EU. Although the behavior of STEC in matrices other than beef has not been extensively studied so far, raw pork products can also act as a vector for STEC transmission to humans. Hence, this study aims to evaluate the safety of ground pork and the dynamics of STEC growth in this material using mathematical predictive models.

Methods:

Model experiments monitored the growth dynamics of STEC in ground pork at various storage temperatures (from 2 °C to 48 °C with a step of 2-3 °C). A mixed suspension of glucuronidase-positive STEC collection strains was used for inoculation. STEC counts were determined by the plate method (ISO 16649-2/2003). Samples for STEC counts were collected and examined immediately after inoculation (0 h) and then at regular time intervals throughout the storage period (48 h to 14 days, depending on the temperature used). The experiment was performed in 3 replicates and a negative control for each temperature monitored. The obtained data were statistically processed and appropriate primary and secondary mathematical models were applied.

Results:

The growth models developed within this study allowed us to evaluate of the effect of temperature on the kinetic parameters of STEC growth in minced pork, to predict the minimum and maximum temperature for STEC growth, to estimate the potential risk to consumers (i.e., to determine the time required to reach the expected infectious dose), and to find out, which models perform best in describing STEC growth in raw minced meat.

Conclusion:

Results of this study broaden the scope of information on growth dynamics of glucuronidase-positive STEC in ground pork as a function of storage conditions. The application of predictive mathematical models to the results obtained using real products (minced pork) provides comprehensive information on the kinetic parameters of STEC growth, thus allowing the estimation of the growth threshold temperature and assessment of potential risk posed by this product to the consumer.

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Salmonella single cell inactivation behavior under acidic food preservation stress

Antonia Angou^{1,2}, Styliani Dimitra Papagianeli², Konstantinos Koutsoumanis², Anagnostis Argiriou^{3,1}, Zafeiro Aspidou^{1,2}

¹Institute of Applied Biosciences, CERTH, Greece, ²Laboratory of Food Microbiology and Hygiene, School of Agriculture, AUTH, Greece, ³Department of Food Science and Nutrition, University of the Aegean

Aim: Food safety is a fundamental issue for consumers and food industry and, lately, the need for a new consideration for the preservation techniques targeting on high quality and mild processing is highlighted. Thus, low pH stress becomes one of the most valuable tools for pathogens control. There is a long list of products which rely solely or to some extent on low pH stress to ensure their safety. The knowledge of only the average population decline is unlikely to be a sufficient basis for processing design and, most importantly, for pathogens such as *Salmonella* with low infectious doses. The aim of this study was to investigate the inactivation behavior of *Salmonella enterica ser. Agona* under acidic food preservation stress focusing on addressing low pH single cell inactivation as well as its implication on population dynamics but also the mechanisms underlining single cell behavior.

Methods: A direct microscopic time lapse method was developed, using appropriate staining for cell viability. *Salmonella* cells were exposed to various acidic conditions and the actual inactivation times of the cells in a population were estimated.

Results: The time of inactivation of single cells was highly heterogeneous. Individual cell inactivation times were fitted to a variety of continuous distributions. The best fitted distribution was further used to predict the inactivation of *Salmonella* populations of various initial levels using Monte Carlo simulation. The simulation results showed that the variability in inactivation kinetics is negligible for concentrations down to 100 cells. As the concentration decreases below 100 cells, the variability increases significantly indicating that the traditional *D*-value used in deterministic first order kinetic models is not valid.

Conclusion: The direct assessment of individual cell inactivation behavior has the potential to increase the accuracy in risk assessment models as well as to be the basis of stochastic microbial inactivation models for the development and improvement of risk-based designs and food safety management systems.

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Single-cell antibiotic resistance analysis: Moving towards a probabilistic assessment of Minimum Inhibitory Concentration (MIC)

Styliani Dimitra Papagianeli¹, Euterpi-Marina Benvenuto¹, Leonardos Stathas¹, Zafeiro Aspidou², Konstantinos Koutsoumanis¹

¹Aristotle University Of Thessaloniki, ²Institute of Applied Biosciences, Centre for Research and Technology Hellas (CERTH)

Aim: Resistance of pathogenic bacteria to antibiotics poses a worldwide threat to food safety and human health, due to their widespread and often inappropriate use. Until now the majority of the studies for the estimation of Minimum Inhibitory Concentration (MIC) of various antimicrobials is conducted with high population levels and by considering microbial populations as a whole. However, single cells of isogenic bacterial populations are characterized by behavioral heterogeneity of phenotypic origin under various environmental conditions thus, it can be expected that single cell behavior is heterogeneous at the presence of antibiotics. Consequently, the objective of this study was to assess single cell behavior at the presence of antibiotics and the implications for the population growth dynamics.

Method: The growth of four *Salmonella enterica* strains on solid media with different concentrations of the antibiotic ampicillin was studied. For this, a modified agar dilution method was developed, where the incorporation of varying concentrations of the antimicrobial agent into the agar medium, using not only serial two-fold dilutions but also the intermediate ones, was followed by the inoculation of different microbial inoculum levels varying from 10^2 to 10^4 CFU/ml onto the agar plate surface.

Results: Considering that every bacterial colony originates from a single cell, a heterogeneous behavior of *S. enterica* single cells of the different strains was observed. The presence of antibiotics affected the probability of growth, which can further be translated to individual cell MIC. To interpret the observations, the variability of the probability of growth was characterized using an appropriate probability distribution and through Monte Carlo simulation the effect of the inoculum level on the MIC, as well as its probabilistic nature were illustrated.

Conclusion: The findings of the present study can outline the resultant of the population behavior at various initial population levels under the exposure to various antibiotic concentrations and provide a new perspective to MIC estimation methodologies through a probabilistic approach.

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Modelling the inactivation of dry-adapted *Cronobacter sakazakii* during the domestic preparation of infant formula

Mr. Enrico Pavoni¹, PhD Elena Dalzini¹, PhD Elena Cosciani-Cunico¹, Paola Monastero¹, Daniela Marigo¹, Stefania Ducoli¹, Alessandro Norton¹, Marina-Nadia Losio¹

¹Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna "Bruno Ubertini"

Aim:

Cronobacter sakazakii (Cr) can survive in dry foods like powdered infant formula and can be life-threatening for immunodepressed or under two months old infants. This study aimed to develop predictive tools to evaluate the Cr inactivation in powdered milk using dry-adapted inoculum to consider the increasing thermal resistance of Cr.

Method:

Strain selection: K_{max} and D-value were evaluated on 7 strains (registered and wildtype) at 55°C using GInaFIT software (Geeraerd et al., 2005). Increasing thermal resistance: K_{max} and D-value were evaluated for a single strain at 55°C using fresh or dry-adapted- inoculum (freeze-dried strain maintained for 14 days at room temperature). Thermal death time curves: z-value was calculated by regression of log D-value calculated at 55, 60, 63°C. Modeling the inactivation during the microwave heating: a central composite design was implemented to study the effect of 3 factors: power (P), treatment time (t) and milk volume (V) and considering five levels for each. Model validation: inactivation of Cr was tested in 2 different microwaves and accuracy factor (Af) and bias factor (Bf) were calculated according to Baranyi et al., 1999.

Results:

ATCC 29544 Cr strain showed the highest D-value of 5.3 ± 0.1 and 9.1 ± 0.5 min in selective and non-selective media respectively, and increasing thermal resistance of dry-adapted- inoculum (D-values of 9.5 ± 1.57 and 12.5 ± 0.1 min, respectively). The z value was 9.7°C and it could be used to determine time-temperature combinations giving a specified bacterial inactivation. The model in microwave heating was: $\text{Inactivation} = -4.4531 + 0.10 * V - 0.0018 * P - 1.9254 * t - 0.0005 * V^2 + 0.000063 * V * P + 0.000009 * P^2$ (ES 0.47) with Af:1.88 and Bf:1.35. Response surfaces were drawn to illustrate the effect of factors on Cr inactivation.

Conclusion:

The results showed the increasing thermal resistance of Cr due to dry-adaptation, implementing the quantitative risk assessment of Cr in powdered milk. They can help manufacturers to improve label information on commercial formulas too.

Antibiotic resistance of *E. coli* isolated from conventional and antibiotic free meat chicken production

Patrícia Melo¹, Emanoelli dos Santos¹, Evelyn da Silva¹, Gabriella Cazolda¹, Gean Carlo Azinari¹, Bruna da Silva², Wanderson Teixeira¹, Prof. Dr. João Pessoa Araújo Junior², Prof. Dr. Carlo Spanu³, Prof. Dr. Fábio Possebon², **Dr. Juliano Pereira**¹

¹São Paulo State University (UNESP), School of Veterinary Medicine and Animal Science, ²São Paulo State University (UNESP), Institute of Biotechnology, ³Università degli Studi di Sassari (UNISS), Department of Veterinary Medicine

Aim:

Foodborne diseases and antimicrobial resistance are a significant threat to public health and the poultry production chain has a significant role in the shedding of antimicrobial resistance. The aim of this study was to evaluate the antibiotic resistance profile of *Escherichia coli* isolated from conventional and antibiotic-free meat chicken farms in Brazil.

Method:

Poultry and farm samples were collected from eight chicken producers (four from conventional system and four from antibiotic-free system). For each farm, the production cycle was accompanied, with samples collected before the housing and during the chicken's production, as well as during the batch slaughter. In each farm a 68 samples were collected at different points during the production cycle (water, feed, insects, drag swabs, cloaca swabs and farm environment) and during slaughtering (slaughterhouse swabs, carcasses, and cecum), for a total of 544 samples. The samples were submitted to *E. coli* detection (ISO 16649) with molecular confirmation (*uspA* gene). Isolates were submitted to antibiotic resistance by CLSI disk diffusion (12 antimicrobial agents from 9 classes). Isolates resistant to three or more classes were considered multidrug-resistant (MDR).

Results:

A total of 504 *E. coli* isolates were obtained, 256 from the conventional and 248 from the antibiotic-free production system. From conventional production, 255 (99.6%) isolates showed resistance to at least one antibiotic and 250 (97.6%) were classified as MDR. In contrast, 217 (87.5%) isolates from antibiotic-free production showed resistance to at least one tested antibiotic and 168 (67.7%) were classified as MDR *E. coli*. When comparing the production systems there was a significant association between conventional production and high frequency of MDR *E. coli* (χ^2 79.6536; $p < .05$).

Conclusion:

The results indicate a high frequency of MDR *E. coli* in samples from conventional production suggesting that the type of production seem to influence in the antibiotic resistance, but a considerable occurrence of MDR strains was also present in antibiotic free system. This finding represents an important global health issue from the one health perspective due to the possible spread of MDR to nosocomial pathogens. The authors would like to thank FAPESP for funding this study (Process 22/03062-6; 23/01185-6 and 23/01195-1).

TemeRe: An online stochastic quantitative microbiology tool for risk-based decision-support in the Food Industry

Mr. Constantine Richard Stefanou¹, Prof. Konstantinos Koutsoumanis¹

¹Laboratory of Food Microbiology and Hygiene, Department of Food Science and Technology, School of Agriculture, Faculty of Agriculture, Forestry and Natural Environment, Aristotle University of Thessaloniki

Aim: This study aimed to develop an easy-to-use web application that integrates stochasticity into the ComBase broth growth models. The focus is on improving user experience and increasing the user pool of stochastic quantitative microbiology tools in the food industry and allowing for shelf life estimations complimentary to challenge testing methods.

Method: Utilising R and the Shiny package, we developed a web application with a graphical user interface to introduce stochasticity in the ComBase broth growth models for 14 bacteria species associated with foods. Stochasticity was introduced into the models by allowing users to define probability distributions in a interactive way for the model inputs, to account for variability in parameters (Initial concentration, temperature, pH, a_w , antimicrobial concentrations and time). Additionally the uncertainty around the estimated maximum growth rate was also included. The user can perform Monte-Carlo analysis for up to 1,000,000 iterations and view the resulting histograms for sampled inputs and predicted final bacterial concentrations. The app also provides the percentage of units exceeding a user-set limit and can allow for the calculation of the shelf-life that complies with a user defined target percentage for non compliant units within set confidence intervals. A case study was carried out for *Bacillus cereus* in milk.

Results: Users can easily quantify the inherent variability in growth parameters, via the UI for *Bacillus cereus* in milk, perform Monte-Carlo analysis and receive quick results for more informed risk-based growth estimations for decision-making in the food industry. The shelf-life option provides a complimentary tool for date marking, reducing the number of needed challenge tests.

Conclusion: The applications can help bridge the gap between research in stochastic microbiological models and practical industrial applications in food safety and quality assurance, improving risk management practices in the food industry, by enabling users to make data-driven decisions to assist in reducing consumer health risks and minimising food waste due to microbiological spoilage.

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An automated data-driven decision support tool to evaluate microbiological and chemical safety of food products

Dr Hermien van Bokhorst-van de Veen¹, Dr. Masja Nierop Groot¹, Dr. Bengü Öztürk¹, Dr. Esther van Asselt², Dr. Hasmik Hayrapetyan¹

¹Wageningen Food & Biobased Research, ²Wageningen Food Safety Research

Aim:

Microbiological and chemical contaminants can be a major threat for the safety, stability, and quality of food products. The aim of the presented project is to provide food producers with a data-driven decision support tool for hazard identification. The tool can be used to (re)design production processes or to make new product formulations, including ingredients from plant-derived side streams.

Method:

The hazard identification tool combines existing data retrieved from scientific literature on potential microbiological and chemical hazards of food categories. For plant-derived side streams the hazards were identified based on the production process, combined with existing data about the commodity. The expert opinion was taken into account. Ingredients in the tool have a strong focus on plant-based. Knowledge rules were formulated to assess the impact of processing steps, such as heating and storage. For the processes where the effect was difficult to assess, a warning text was formulated.

Results:

The hazard identification tool is web-based and qualitative. A total of 29 food ingredient categories and 10 plant-derived side streams were included and can be selected to make a food formulation simulation. The impact of 19 processing steps, such as heating and storage, on microbiological and chemical hazards was evaluated based on literature, and either a knowledge rule or a warning text was formulated. The knowledge rules allow to assess the impact of freezing, high hydrostatic pressure, heating and storage on microbiological hazards and the impact of heat on chemical hazards. The user can select the order of processing steps applied. The outcome of the tool is a list of identified microbial and chemical hazards for the food product formulated by the user. The tool will be demonstrated with a plant-based burger case.

Conclusion:

To ensure the highest quality and safety, production processes need continuous assessment of hazards and control strategies. This is especially important if changes occur in the process. The outcome of the tool can be used as an input for the HACCP plan. This will help food business operators to assess the product safety at an early stage of product development in a fast and reproducible way.

Resilient food safety management to anticipate on related microbiological food safety challenges

Mathis Vermeersch¹, Mieke Uyttendaele¹, Liesbeth Jacxsens¹

¹Ghent University

Aim: Food businesses are operating in a rapidly changing external business environment, for example, the growing population, climate change, the covid-19 crisis, etc., impacting local and global markets. The external business environment is hosting drivers, being social, technological, environmental, economic and political (STEEP), directly impacting microbiological food safety, thus the business its food safety management system. Understanding (i) the drivers for the emergence of microbiological food safety risks, and (ii) their impact on food safety management system activities, enables appropriate resilience building in food businesses.

Method: A scoping literature review, focussing on known microbiological hazards was carried out. Through inductive coding, bringing together the established scientific literature (scoping review) and grey literature, drivers for the emergence of microbiological food safety risks were explored. An expert workshop with food business operators was organised in Belgium to assess the impact of the identified drivers on selected food safety management activities (quality control, quality assurance and food safety performance).

Results: Drivers for the emergence of microbiological food safety risks have been identified, e.g. consumer behaviour, global trade, and climate change. A qualitative, hazard-focused analysis elucidates major drivers affecting the occurrence or emergence of specific microbiological hazards. From the expert workshop, (i) major drivers impacting internal food safety management activities were identified, together with (ii) the mechanism of impact, as input to the design of resilience building blocks in future-proof food safety management.

Conclusion: This research brings together drivers for the occurrence of known microbiological hazards in the European food system. The impact of the identified drivers on the level of food business operators their food safety management system was assessed to gain understanding in resilience building blocks for future-proof food safety management. The output of this research will be used in the development of a diagnostic tool to assess the maturity and resilience of food businesses their food safety management system.

Combined Application of Recombinant Holin and Endolysin Against *Salmonella* Kentucky

Student Segah Yetiskin¹, Yeşim Soyer

¹Middle East Technical University

Aim:

Salmonella is a significant global health concern due to its role in foodborne illnesses. World Health Organization (WHO) states that the increase in resistance of *Salmonella* to antibiotics poses a threat. The resistance of pathogens to both antibiotics and bacteriophages, which are presented as an alternative to antibiotic resistance, has increased the tendency towards bacteriophage-derived proteins. This study aims to explore the use of bacteriophage-derived enzymes, specifically endolysins and holin proteins, to target and lyse *Salmonella* cells effectively, offering a potential alternative to traditional antibiotics and chemical treatments.

Method:

The study utilized endolysin LysSK137 and holin protein HlnSK137, both derived from previously sequenced phage genomes. The proteins were transferred to *E. coli* BL21 cells using recombinant DNA technology and purified using a His-Tag purification kit. Bioinformatics tools were employed to analyze the structure and properties of HlnSK137. The lytic activity of these proteins, alone and in combination, was tested against *Salmonella* Kentucky to evaluate their effectiveness in penetrating bacterial cell walls and causing cell lysis.

Results:

Bioinformatics analysis revealed that HlnSK137 is an unstable protein with a molecular weight of 24.7 kDa and an isoelectric point of 8.40. The lytic activity tests showed that the combined use of recombinant HlnSK137 and LysSK137 caused approximately a 2 log CFU/mL decrease in *Salmonella* Kentucky cell counts. HlnSK137 improved cell wall permeability, enhancing the effectiveness of LysSK137 in entering the cells and executing its lytic functions.

Conclusion:

The findings suggest that the combined application of recombinant HlnSK137 and LysSK137 is effective in reducing *Salmonella* populations, with potential applications in food safety, health, and veterinary medicine. HlnSK137 could serve as a non-chemical alternative for improving cell wall permeability, which could be further optimized for use with LysSK137 to enhance its antibacterial efficacy.

Unveiling c-di-AMP and PdeA Gene in *L. monocytogenes*: Acid and Oxidative Stress Resistance

Mahide Muge Yilmaz Topcam¹, Dimitris Balagiannis¹, Kimon Andreas Karatzas¹

¹University of Reading

Aim: Bacterial infections cause serious problems in animals and humans, highlighting the need for a comprehensive grasp of bacterial adaption within the host environment. c-di-AMP (cyclic-di-adenosine monophosphate) is a signalling molecule and is critical for bacterial adaptation and pathogenesis by controlling essential physiological processes crucial for survival and disease development. In this study, we elucidate the multifaceted role of c-di-AMP in *Listeria monocytogenes*, focusing on its impact on bacterial growth, heat resistance, biofilm formation, and response under acid and oxidative stress.

Method: *L. monocytogenes* 10403S WT and *pdeA* deleted mutant were used in the experiments. Cells were grown in BHI until the stationary phase and challenged with heat (56°C for 30min), acid (1M HCl for 1h) and oxidative stress (30% H₂O₂ for 1h) and, survival rates were detected for resistance experiments. Under the same conditions, c-di-AMP levels were measured by using LC-MS to investigate the effect of c-di-AMP concentrations on the resistance mechanisms. Biofilm formation was measured in BHI at determined time intervals by using the crystal violet method.

Results: We discovered that deleting the gene for PdeA doesn't affect the bacteria's short-term survival when subjected to acidic shock conditions. The extracellular c-di-AMP supplementation helps to restore changes in the levels of intracellular GABA. We also found that mutant bacteria $\Delta pdeA$ are more sensitive to oxidative stress compared to normal bacteria. This sensitivity is to be related to the levels of c-di-AMP inside the cells. c-di-AMP concentrations changed in different trends depending on the applied stress conditions. We showed that deletion of the *pdeA* gene didn't significantly affect the biofilm formation at neutral pH value but in acidic conditions.

Conclusion: c-di-AMP is needed for the efficient functioning of a system related to GABA during acid stress. In this study, we explained the possible connection between c-di-AMP concentrations and the GAD system under acid shock. The change in c-di-AMP levels under different stress conditions points to the effect of different mechanisms on c-di-AMP secretion and degradation. Our study provides new insights into how bacteria like *L. monocytogenes* respond to stress, highlighting the importance of c-di-AMP in their adaptation to different environmental challenges.

Spore germination and growth of *Bacillus cereus* in hot tomato sauce

Mgr. Phd. Alena Zouharová¹, Marta Dušková¹, Michaela Čutová¹, Josef Kameník¹

¹University Of Veterinary Sciences Brno

Aim:

Bacillus cereus is highly efficient in spore formation, and can form spores with high heat resistance. The aim of this study is to determine the conditions (temperature and time) of *Bacillus cereus* growth in hot tomato sauce with different pH after its artificial contamination by spore suspension.

Methods:

The bacterial suspension of selected *Bacillus cereus* strains (CCM 869; DSM 4312) was incubated aerobically at 30 °C for 10 days. The homogeneous spore suspension was washed and treated with ultrasonication to inactivate any vegetative cells. The spore suspension thus prepared was used to artificially contaminate tomato sauce (pH 4,4 and 6,1) at 70 °C. The contaminated tomato sauce was packed in 150 g portions and stored at 40 °C, 50 °C, 60 °C. The samples were examined immediately after packaging and after 1, 2, 3, 4 hours of storage at the given temperatures. *Bacillus cereus* was cultured on MYP agar at 30 °C/24 h/aerobically.

Results:

Spores of strain DSM 4312 germinated in high numbers, especially in hot tomato sauce with adjusted pH 6.1 (3 log CFU immediately after packaging). In tomato sauce with pH 4.4, spore germination occurred less (2 log CFU immediately after packaging). Strain CCM 869 did not grow well in either type of sauce (1 log CFU throughout incubation). During storage of packaged food at 40 °C, the highest increase in *Bacillus cereus* counts occurred after 2 to 3 hours of storage, up to 5 log CFU in the tomato sauce at pH 6.1 and 3 log CFU at pH 4.4. During storage at 50 °C and 60 °C, the highest counts were also observed between hours 2 and 3, but the counts were lower (2 log CFU).

Conclusion:

In our study, a significant difference was found in the germination and growth characteristics of the strains used. Furthermore, pH was found to be a significant selection factor in food as well as the temperature at which hot food is stored or transported. A storage time of 2-3 hours at 40 °C was found to be the most hazardous in terms of food safety. The results will allow the definition of good distribution practice principles for the distribution of ready meals, which will contribute to improved food safety and to the reduction of food waste.

Acknowledgment:

This study was supported by the grant NAZV QK23020061.

EFSA's Risk Assessment and characterisation of microorganisms in the food chain

Dr. Yolanda Garcia Cazorla¹

¹European Food Safety Authority (EFSA)

Aim: Ensuring safety and efficacy of feed additives containing/produced with microorganisms

In the European Union (EU), the introduction of feed additives into the market requires authorisation under Regulation (EC) No 1831/2003. The European Food Safety Authority (EFSA) is responsible of assessing the efficacy and safety of these additives for animals, humans and the environment. Based on EFSA's assessment, the European Commission will grant or deny the authorisation in the EU market. Microorganisms used in or to produce feed additives undergo rigorous risk assessments, with EFSA establishing comprehensive data requirements to facilitate evaluation and decision-making.

Method: Scientific Risk Assessment

EFSA conducts scientific risk assessments based on technical dossiers submitted by applicants. These dossiers provide crucial data necessary for safety and efficacy evaluation. For additives containing microorganisms or obtained/produced from/with microorganisms, characterisation of the microorganisms provide information relevant for the assessment. The microorganisms should be characterised for their taxonomic identification and presence of traits/genes of concern. For genetically modified strains, EFSA characterises the genetic modifications and determines its safety. Additionally, additives derived from microorganisms need to be characterised for aspects related to the strain(s) from which they are prepared/obtained from/with (e.g., antimicrobial activity, presence of viable cells and/or DNA).

Results: Scientific basis for EU Regulations

EFSA's assessments culminate in scientific opinions that serve as a cornerstone for regulatory decision-making in the EU. These opinions provide vital insights into the safety and efficacy of feed additives, enabling regulators to make informed decisions regarding market authorisation. EFSA ensures transparency by making all scientific opinions accessible to the public through its website, fostering trust and accountability in the regulatory process.

Conclusion: Ensuring a safe and transparent food supply chain

EFSA's pivotal role in safeguarding the EU's food supply chain involves providing rigorous scientific assessments to EU risk managers. This contribution helps uphold high standards of food safety and protection for humans, animals, and the environment. Recognizing the paramount importance of regulated products, EFSA prioritises the development of guidance documents outlining requirements. Furthermore, EFSA is actively enhancing and streamlining its processes, underscoring its dedication to ensuring a safe and transparent food supply chain for all stakeholders.

Biofilms: Mechanical and Morphological Investigation

Mrunmayee Joshi¹, Laura Buccoli¹, Michael Milward², Peter J Fryer¹, Zhenyu Jason Zhang¹

¹School of Chemical Engineering, University Of Birmingham, ²School of Dentistry, University of Birmingham

Aim:

In the food industry, biofouling occurs when microorganisms form an unwanted layer (biofilms) on food processing surfaces. Biofouling can lead to reduced heat flow along with high energy and product loss. In addition to reducing process efficiency, they can cause cross contamination and spoilage.

Extensive microbiological investigations have been carried out in evaluating biofilms, whilst the knowledge and capability to characterize the mechanical properties and hence improved cleaning strategies are limited. This work develops a new strategy to generate biofilms of *S. mutans* consistently and investigating their physical properties.

Method:

A customised high-throughput setup was established, where a glass container was modified to hold substrates vertically in an oxygen-deficient environment. The container was then placed in an orbital shaker at 37 °C for 6 or 9 days with media replaced every 3 days. Biofilms were grown in media containing either 1 % (HS) or 0.1 % (LS) sucrose solution. The developed biofilms were stained with SYTO 9 and Propidium Iodide and visualised using Confocal Laser Scanning Microscopy (CLSM). A home-built microindentation device was used to calculate Young's Moduli which were related to the stiffness with a spherical indenter and 2 mN of threshold force.

Results:

Biofilms generated were assumed to be thin films for the indentation experiments. Results show a range of Young's moduli (0.64 - 0.71 kPa) across different growth conditions (HS and LS) of *S. mutans* biofilm likely explained by variations in nutrient availability for bacterial growth. Differences in morphology were also observed in the two biofilms when CLSM images were compared for different number of growth days (6 or 9).

Conclusion:

This approach will prove useful in understanding how model biofilms can be developed and used to establish an in-depth understanding of mechanical properties for different applications.

Evaluation of nitrite reduction on the microbial safety of cooked smoked pork chouriço

Ms Maria Nunes¹, Inês Cruz², Lúcia Noronha³, Norton Komora¹, Rui Pereira⁴, Joana Barbosa¹, Fátima Carvalho², Paula Teixeira¹

¹Escola Superior De Biotecnologia, ²Primor Charcutaria Prima, S.A., ³Colab4Food, ⁴Tecmeat

Aim:

Nitrite is a preservative used worldwide, particularly in the meat industry, to guarantee certain organoleptic and microbiological properties of delicatessen meat products, acting as a hurdle to pathogenic bacteria and protecting against bacterial spoilage. Cooked smoked pork *chouriço* contains nitrite in its formulation, which raises some concerns regarding the consumer's health, and therefore the European legislation on the use of nitrites has been modified, where the maximum limit of 150 mg/kg of meat has been reduced to 80 mg/kg of meat in the EU. The present study focuses on the assessment of the impact of nitrite reduction on the microbiological safety and organoleptic properties of pork *chouriço*.

Method:

The determination of total viable counts, lactic acid bacteria, *Enterobacteriaceae*, *Escherichia coli* and pathogenic bacteria (*Listeria monocytogenes*, *Salmonella* and spores of sulfite-reducing *Clostridium*) was performed according to ISO standards in pork *chouriço* produced with different nitrite concentrations. Organoleptic characteristics, such as colour, a_w , pH and texture were evaluated at times 0 days, 30 days and 60 days of storage under vacuum at room temperature.

Results:

Although the nitrite concentration has been reduced to almost half of the usual concentration, the microbiological results show that the product remains stable in 60 days of vacuum storage at room temperature and complies with the legal limits, below $1.0E+05$ CFU/g for total viable counts and lactic acid bacteria. Regarding organoleptic properties, due to the heterogeneity of the product, the results show some high standard deviation, especially in colour and texture, but both a_w and pH are in line with the standard values obtained in the products.

Conclusion:

In conclusion, although further tests are needed to establish safety, no significant differences were observed in microbial growth during the product's shelf-life, or in its organoleptic properties. Therefore, there is no evidence of safety problems associated with the consumption of smoked cooked pork *chouriço* with a decrease of more than 45% in the nitrite concentration. However, caution should be taken until further research confirms its safety.

The role of *Chryseobacterium* species in food spoilage

Dr Arina Hitzeroth¹, Mickayla Zuerina¹

¹University Of The Western Cape

Aim:

The genus *Chryseobacterium*, a Gram-negative, heterotrophic, aerobic rod that possesses proteolytic and lipolytic enzyme activity and their psychrophilic nature, isolates from the genus have demonstrated food-spoilage characteristics. Food spoilage of milk, fish, meat, and poultry products has been attributed to *Chryseobacterium* isolates. We tested the ability of chryseobacterial species isolated from fruit environments to cause spoilage of fruit, and we evaluated the co-cultivation with *Pseudomonas aeruginosa* by comparing their carbon utilizations as well as biofilm formation between the organisms.

Method:

Chryseobacterium were cultivated in fruit juice for 48 hours, and the volatile organic compounds were determined with LCMS to determine spoilage. Two *Chryseobacterium* strains, CFS2 and CFS15, were cocultured with *P. aeruginosa* on BIOLOG™ Eco-plates to determine carbon utilisation. The different strains' abilities to produce biofilms have also been assessed by cultivation on different media and different staining techniques.

Results:

In dual-culture growth experiments, the chryseobacterial growth was affected when grown in co-culture, but *P. aeruginosa* growth was not affected. In the EcoPlate™ analysis, the Substrate average well-colour development (SAWCD) analysis indicated that the metabolism of each substrate group was significantly different for single species and co-cultures. The planktonic growth assays revealed that minimal media results in stronger biofilms. *Chryseobacterium* isolates (CFS2, CFS15) preferred RPMI-1640 media for biofilm formation. However, no attachment was observed in any media, and there was a notable increase in biofilm formation in the co-cultures. Volatile organic compound analysis indicated the production of various compounds in both single and co-cultures.

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Conclusion:

The relationship between *Chryseobacterium* and *Pseudomonas* is negative, as chryseobacterial growth was severely affected by *Pseudomonas*. This was observed when comparing carbon source utilisations. They do, however, cooperate in biofilm production, as the planktonic growth showed an improvement in biofilm formation with branchlike structures forming compared to the monolayer biofilm format in a single culture. *Chryseobacterium* seems to favour RPMI media but does not form any attachments in any media. All strains were also able to spoil fruit juice, indicated by various volatile organic compound formations.

Antimicrobial properties of chitosan edible coating with *P. sidoides* extracts against food spoilage organisms

Dr Arina Hitzeroth¹, **Wendy Pretorius**¹

¹University Of The Western Cape

Aim:

The increasing interest in sustainable and eco-friendly methods of food preservation has prompted researchers to explore the use of plant antimicrobials as an alternative to synthetic preservatives. Studies have demonstrated that plant extracts can be used as biodegradable food packaging. Plant extracts, possess the same functional properties as synthetic additives but are environmentally friendly. This study aims to investigate the potential applications of *Pelargonium sidoides*, a South African herb commonly used in the pharmaceutical industry. Additionally, the study examines the impact of chitosan edible coating combined with the medicinal root extracts from *Pelargonium sidoides* on antimicrobial activity against various food spoilage organisms.

Method:

Ethanollic root extracts of *Pelargonium sidoides* were prepared and assessed for their antimicrobial activity against various bacterial species associated with food spoilage and food pathogens using the dilution method on a solid agar medium. The minimum inhibitory concentrations were determined against both Gram-positive and Gram-negative organisms. Fungal and bacterial endophytes were isolated, and the antimicrobial activity was tested and compared to the root extract. A pelargonium-chitosan antimicrobial coating was tested for its antimicrobial properties against food spoilage bacteria through agar diffusion tests, and micro-dilution assays, respectively.

Results:

The *P. sidoides* plant extracts had antimicrobial activity against both Gram-negative and Gram-positive organisms. Fungal endophytes had greater antimicrobial activity than bacterial endophytes. The pelargonium-chitosan indicated some antimicrobial activity but lower than the concentration of the endophyte and the root extract.

Conclusion:

The antimicrobial properties of *P. sidoides* have been known for a long time and have been actively used in pharmaceuticals. In this project, we applied *P. sidoides* to food applications. Further research is required to identify the specific compound responsible for these activities. This will enable the use of the plant extracts as natural components of multi-barrier food preservation systems. The research indicates the synergistic or cumulative effects exhibited by plant antimicrobials, as well as the effective incorporation of plant extracts with food technologies that provide an enhanced hurdle effect, which can improve food safety and extend shelf life.

What is that smell, can you tell?

Joëlle Housmans¹, Imca Sampers¹

¹Research group VEG-i-TEC. Department of Food Technology, Safety and Health (BW23). Faculty of Bioscience Engineering. UGent - Campus Kortrijk

Aim: Within the potato industry, the blanching process is a highly water-intensive procedure. To ensure more sustainable practices, several potato fries companies reuse their blanching water for multiple cycles. However, over time, this reused water produces an undesired odor, adversely affecting the flavor profile of baked potato fries. Our primary aim is to identify and detect the chemical compound(s) responsible for this odor and its origin. Subsequently, we aim to validate an in-line sensor capable of real-time detection, allowing fast and accurate adjustments to the process to ensure both healthy and tasty potato fries.

Method: We performed a comprehensive literature review on previously conducted research concerning potato blanching water, focusing on the presence of microorganisms, spores, thermophiles and associated odor.

Results: Our thorough search revealed insufficient evidence to pinpoint a particular compound responsible for the observed odor. We hypothesize two potential sources for the origin of the odor; a) the potato itself, which as a consequence of heating can leach volatile organic compounds (VOCs) or produce new volatile metabolites, or b) micro-organisms (thermophiles, spores or biofilm-forming), which survive the high blanching temperatures and may breakdown potato-derived compounds into VOCs. Therefore, we listed microorganisms and VOCs commonly associated with potatoes, alongside existing and emerging methodologies for their detection, both in offline and in-line contexts.

Conclusion: Given the knowledge gap about potato blanching water, we set up an elaborate framework providing lists of potential VOCs, microorganisms and detection methods. This information will be disseminated in review format and serve as the basis for future research approaches.

Risk assessment and prediction models for fungal spoilage in strawberries post-harvest

Dr Alessandra Marcon Gasperini^{1,2}, **Vasilis P. Valdramidis**^{1,3}

¹Department of Food Sciences & Nutrition, Faculty of Health Sciences,, ²Clinical, Pharmaceutical and Biological Sciences, School of Life and Medical Sciences, MycoLab, University of Hertfordshire,

³Laboratory of Food Chemistry, Department of Chemistry, National and Kapodistrian University of Athens

Aim: Fungal spoilage in strawberries poses economic risks to growers and distributors, along with the potential for mycotoxin contamination, threatening food safety. In this research, we seek to develop processed soft fruits with improved quality, nutrition, and shelf-life, while also addressing fungal contamination and mycotoxin production risks. Thus, we aimed to identify the most mycotoxin contaminants in strawberries and to construct a comparative risk assessment system for evaluating and ranking fungal and mycotoxin hazards.

Method: Published datasets and peer-reviewed studies were used as representative references (n=71). Fungal contaminants in strawberries were clustered by species, and mycotoxin concentrations were standardized ($\mu\text{g.kg}^{-1}$) and ranked for comparison. Primary models were used to estimate the growth rates (μ) and lag phases (λ) for *Alternaria*, *Penicillium* and *Aspergillus*, with adjustment coefficients (k) for field conditions. A prediction model for post-harvest infection elapsed 7 days (5 storage, 2 retail), considering storage and retail temperatures. Visible spoilage was defined by a 2 mm colony size threshold, with associated risk levels (high, moderate, low) based on species-specific probabilities of spoilage.

Results: The study has shown a high prevalence of *Alternaria* spp. and its relative mycotoxins (alternariol monomethyl ether and alternariol), followed by *Aspergillus*, *Penicillium* spp. Mycotoxins with regulatory limits (*i.e.*, fumonisin, aflatoxins, ochratoxin and patulin) were largely reported in strawberries. Risk assessment models predicted a low risk of spoilage when strawberries were stored and retailed at 4°C. However, increasing the retail storage temperature to 15°C resulted in an increased risk of spoilage by *Alternaria* spp. Additionally, a scenario with storage at 4°C and retailing at 25°C predicted a high risk of spoilage by *Alternaria* spp. and moderate spoilage risk by *Aspergillus* and *Penicillium* spp.

Conclusion: *Alternaria*, *Aspergillus* and *Penicillium* spp. were the most common spoilers in strawberries, with associated mycotoxins posing risks. Temperature control is crucial to mitigate spoilage, but species-specific growth factors must be integrated into risk assessments. Future research will assess mycotoxin risk to build model-based process optimization for the production of safe and stable products.

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Antibacterial activity of the cassava-derived natural flavor Cloud S-C100 coated films against spoilage-associated bacteria

Ayaka Nakamura¹, Aya Hatanaka¹, Takashi Kuda¹, Hajime Takahashi¹

¹Tokyo University Of Marine Science and Technology

Aim:

“Active packaging” is a packaging technology that actively adds functions to maintain food quality and has been attracting attention in recent years. The quality of food is closely related to the growth of microorganisms in food and controlling them is a constant challenge for the food industry. In this study, the antibacterial properties of a film coated with Cloud S-C100, a natural flavor derived from cassava were evaluated, and whether it could be used as an actual food packaging film were examined.

Method:

First, to clarify the active ingredients with antibacterial properties, a component analysis of Cloud S-C 100 solution using GC-MS was performed. Next, Antibacterial films were created by coating the base film with Cloud S-C 100 solution using a gravure printing machine. Two types of spoilage-related lactic acid bacteria were inoculated onto the surface of the ham at a concentration of 3 log CFU/g, and both sides were sandwiched between antibacterial films. A polyethylene film was used in the control group. It was stored at 4°C for 2 weeks, and the number of lactic acid bacteria was measured on the 7th and 14th days.

Results:

From the results of GC-MS analysis, 58 components including aldehydes, ketones, organic acids, furans, pyrans, and phenols were detected. The highest percentage was 3-Furaldehyde, followed by 1-Hydroxy-2-propane and Hydroxy-acetaldehyde. The results of growth inhibitory test showed that when using the Cloud s-c100 coating film, the growth of lactic acid bacteria was completely inhibited on the 7th day, and even on the 14th day, inhibition of 3log CFU/g was confirmed compared to when using the control film.

Conclusion:

Films coated with Cloud S-C 100 were shown to inhibit the growth of lactic acid bacteria even when used in food products. Active packaging with antibacterial properties is expected to extend the shelf life and ultimately reduce the food loss.

Integrating spoilage modeling in QMRA: *Listeria monocytogenes* and Lactic Acid Bacteria in sliced deli meats

Mr. Constantine Richard Stefanou¹, Mrs. Nikola Smigielska^{1,2}, Prof. Konstantinos Koutsoumanis¹

¹Laboratory of Food Microbiology and Hygiene, Department of Food Science and Technology, School of Agriculture, Faculty of Agriculture, Forestry and Natural Environment, Aristotle University of Thessaloniki, ²Department of Food Quality, prof. Wacław Dabrowski Institute of Agricultural and Food Biotechnology - State Research Institute

Aim: This study aimed to integrate quantitative microbiological risk assessment (QMRA) for a pathogen with quantitative microbiological spoilage risk assessment (QMSRA) for a specific spoilage organism (SSO) into a single combined model to provide a more realistic estimation of foodborne illness risk, considering the variability in consumer perception of spoilage. The combined model was developed for *Listeria monocytogenes* and Lactic Acid Bacteria (LAB) in sliced deli meats in Greece and Poland.

Method: Microbiological testing of deli meat samples and historical data were collected for the prevalence and concentration of the pathogen *Listeria monocytogenes* and the SSO *Lactic Acid Bacteria* (LAB) in prepackaged sliced deli meats from Greece and Poland. Product physicochemical properties (pH, water activity, and nitrites concentration) influencing bacterial growth were analysed. Domestic storage temperature data was collected and consumer behaviour regarding consumption frequency and storage practices were surveyed via questionnaire with n=200 participants. Probability distributions were fitted to the collected data to describe variability of the model inputs. The predictive microbiology model for *Listeria monocytogenes* and LAB in meat products developed by Mejlholm and Dalgaard was used to estimate microbial growth during product storage. A dose-spoilage response model derived from consumer sensory testing, was integrated into the QMRA model, to describe the variability in spoilage perception among consumers. Sensitivity analysis was performed on the final combined model. Distribution fittings, Monte Carlo analysis and sensitivity analysis were performed in Microsoft Excel with the @Risk add-in by Lumivero.

Results: The combined risk assessment model yielded a more realistic estimation of foodborne illness risk. The inclusion of the spoilage module resulted in a lower estimated combined risk for listeriosis, indicating the significance of accounting for spoilage-related factors in risk assessment.

Conclusion: Integrating QMRA and QMSRA methodologies for pathogens and spoilage microorganisms can enhance the accuracy of foodborne illness risk estimation. This approach provides a more holistic understanding of food safety risks, acknowledging the impact of spoilage.

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Thermal inactivation of *Bacillus subtilis* spores and vegetative cells at 60°C under low vacuum condition

Hiyori Taki¹, Kento Koyama¹, Shige Koseki¹

¹Hokkaido University

Aim:

In recent years, low vacuum cookers have emerged as a novel creative cooking procedure to season and cook foodstuffs quickly and mildly. We focused on the reduced boiling point under low vacuum condition, because there might be some effect on bacterial inactivation in a boiling state. The objective of this study was to investigate whether bacterial spores and vegetative cells could be inactivated at low temperature under low vacuum condition.

Method:

Bacillus subtilis spores and vegetative cells (ca. 10⁶ CFU/mL) suspended in distilled water (10 mL) in a glass tube were heated at 60°C under constant atmospheric pressure or low vacuum (90 kPa) for 20 min using a low vacuum cooking apparatus (Vide Pro, EVC-221, ESPEC, Osaka, Japan). In addition, repeated process of depressurization (low vacuum state) and release (atmospheric pressure) was applied to *B. subtilis* spores and vegetative cells at 60°C for 7 cycles. Viable cell numbers before and after each treatment were determined by direct plating method on tryptic soy agar.

Results:

There was no significant reduction in the survival of *B. subtilis* spores regardless of the treatments at 60°C with or without low vacuum. In contrast, there was significant difference in the survival of *B. subtilis* vegetative cells after treatment at 60°C for 20 min between atmospheric pressure and low vacuum conditions. The survival number of *B. subtilis* vegetative cells heated at 60°C under low vacuum condition (1.3 log CFU/mL) was significantly lower than those under atmospheric condition (2.1 log CFU/mL). In addition, repeated process of depressurization (low vacuum state) and release (atmospheric pressure) inactivated *B. subtilis* vegetative cells by 1.0 log CFU/mL. There was no significant difference in the survival *B. subtilis* vegetative cell numbers between constant vacuum condition and the repeated process.

Conclusion:

The results presented in this study suggest that low vacuum environments may facilitate inactivation of bacterial vegetative cells at low temperatures such as 60°C, but not for spores.

Inactivation of *Alicyclobacillus acidoterrestris* spores in functional tomato juice processed by heat and UV-C light

Rose Daphnee Tchonkouang^{1,2}, Maria Dulce Antunes^{1,2}, Maria Margarida Vieira^{1,3}, Mecit Halil Öztop

¹MED-Mediterranean Institute for Agriculture, Environment and Development & CHANGE—Global Change and Sustainability Institute, Faculty of Sciences and Technology, Universidade do Algarve,

²Faculty of Science and Technology, Universidade do Algarve, Campus de Gambelas, 8005-139 Faro, Portugal,

³Department of Food Engineering, High Institute of Engineering, Universidade do Algarve, Campus da Penha, 8005-139 Faro, Portugal

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Tchonkouang, Rose Daphnee^{1,2}; Antunes, Maria Dulce^{1,2}; Vieira, Maria Margarida^{1,3}

¹MED-Mediterranean Institute for Agriculture, Environment and Development & CHANGE—Global Change and Sustainability Institute, Faculty of Sciences and Technology, Universidade do Algarve, Campus de Gambelas, 8005-139 Faro;

²Faculty of Science and Technology, Universidade do Algarve, Campus de Gambelas, 8005-139 Faro;

³Department of Food Engineering, High Institute of Engineering, Universidade do Algarve, Campus da Penha, 8005-139 Faro, Portugal

³Department of Food Engineering, High Institute of Engineering, Universidade do Algarve, Campus da Penha, 8005-139 Faro, Portugal

Acidothermophilic bacteria, particularly *Alicyclobacillus acidoterrestris*, are recognized as key target microorganisms for the quality control of pasteurized acidic foods. Consequently, *A. acidoterrestris* has been proposed as a reference microorganism in designing effective pasteurization protocols for acidic fruit products (Sourri et al, 2020). The effectiveness of continuous UV-C treatment (UV-C dose of 0.44 J/l at a flow rate of 3.3 l/h) and conventional heating in isothermal (85, 90, and 95 °C) and nonisothermal (90 °C at a flow rate of 4.9 l/h) settings for reducing spores of *Alicyclobacillus acidoterrestris* CECT 7094 was investigated in a functional tomato juice. The kinetic parameters (D- and z-values) were determined during conventional isothermal heating. D-values at 85, 90, and 95°C were 95.23, 19.01, and 7.83 min, respectively. No significant differences between continuous UV-C treatment and continuous pasteurization were observed ($P > 0.05$). Results showed significant lethality for spores treated only above 85 °C in conventional isothermal heating. UV-C and nonisothermal (continuous) treatment exhibited a similar reduction in microbial count. Though UV-C treatment can be a cost-effective and eco-friendly alternative to thermal treatment, both pasteurization techniques were ineffective in reaching the required 5 D reduction on *A. acidoterrestris* spores (our goal for food safety in acidic products), as only a 2D reduction was obtained in the time, temperature and UV-C ranges applied. However, further studies combining heat and UV-C can improve the effectiveness and be able to kill heat-resistant spores to achieve the aforementioned goal.

Keywords: *Alicyclobacillus acidoterrestris*, UV-C light, heat, food processing, tomato juice

Acknowledgements

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Reference

Sourri, P., Argyri, A. A., Panagou, E. Z., Nychas, G.-J.E., & Tassou, C. C. (2020). *Alicyclobacillus acidoterrestris* strain variability in the inactivation kinetics of spores in orange juice by temperature-assisted high hydrostatic pressure. *Applied Sciences*, 10(21), 7542.

Antifungal activity of biosurfactants against bread spoilage strains and their development of resistance

Ana Sousa¹, Dr. Konstantina Kourmentza², Dr. Paula Jauregi^{3,4}, Dr. Kimon-Andreas Karatzas¹

¹Department of Food & Nutritional Sciences, University of Reading, ²Faculty of Engineering, University of Nottingham, ³Efficient and Sustainable Processes, AZTI, ⁴IKERBASQUE, Basque Foundation for Science

Aim:

Microbial spoilage of bakery products, primarily caused by fungi, substantially contributes to the food waste global problem. Consumers and public authorities are increasing the demand for natural, less toxic, and sustainable ingredients in food. This study aims to evaluate the efficacy of biosurfactants against fungal bread spoilage strains in accelerated shelf-life tests (ASLTs) on bread and evaluate the evolution of resistance of fungi against these compounds.

Method:

ASLTs were performed, to determine biosurfactants' ability to delay mould growth on bread, when compared to control bread without preservatives and reference bread with calcium propionate (chemical preservative). To achieve this, bread crumb slices were spot inoculated with 100 spores of *A. niger* or *P. paneum* on the surface and incubated at room temperature. Images of the slices were captured every 20 min, to determine the time of appearance (ToA) of moulds. Four different biosurfactants (A-D) were used in ASLTs, in concentrations ranging from 0.1 to 0.6% (flour wt. basis). Additionally, *A. niger* and *S. cerevisiae* were used in assays (in vitro) where they were exposed to increasing concentrations of biosurfactant A, to assess their development of tolerance against this compound. This was done by transferring cells into PDA plates containing concentrations of biosurfactant A corresponding to 10-250% MIC of each strain, until growth was no longer observed.

Results:

In ASLTs, biosurfactant B was the most effective compound delaying appearance of moulds. It was able to outperform calcium propionate at equal concentrations (0.3% flour wt. basis), by significantly delaying the ToA of *A. niger* and *P. paneum*. In development of tolerance assays, both *S. cerevisiae* and *A. niger* were able to acquire tolerance to biosurfactant A up to 200% MIC.

Conclusion:

Biosurfactant A exhibited stronger activity against *A. niger* and *P. paneum* than the reference chemical preservative, when applied to bread, suggesting its potential as a natural alternative in bread preservation. *S. cerevisiae*'s increased tolerance to this compound is a positive outcome for the use of biosurfactant B in yeast-raised goods. Nevertheless, the evolution of resistance of spoilage strains against this compound would be problematic for the bakery industry, and needs to be further investigated.

Combining Culture-Dependent and Culture-Independent Techniques to Secure Paint and Cosmetic Quality

Prof. Dr Katrin Bach¹, Marleen Mohaupt¹

¹MCI | The Entrepreneurial School®

Aim

Consumer demands in terms of quality and ingredients have a major influence on the development of new products in both the paints & coatings and cosmetics sectors. The use of physiologically harmless raw materials, the avoidance of chemically synthesized preservatives and sustainable production processes are becoming increasingly important. The reduction of various additives and the increased use of water-based formulations brings with it problems in terms of shelf life and storage stability due to increased microbial contamination. In order to reduce the microbial load in paint, varnish and cosmetic products and thus extend their shelf life, the manufacturing process and the raw materials involved are to be analyzed microbial and methods, which are already established in the food industry, are to be transferred to the chemical and cosmetics industries.

Method

To achieve these goals, the critical points for microbiological stability in the production areas were identified and subjected to regular monitoring. In addition to the products, this also includes the raw materials, the manufacturing plant and as well the environment of the production itself. The hygiene was described using swab tests and dipslides. The air was sampled using a combination of active and passive air sampling in accordance with ISO standard 16000. The products and production raw materials such as water were tested for their total aerobic mesophilic bacterial count using the plate casting method in accordance with ISO standard 4883. Sanger sequencing and MALDI-TOF were used to qualitatively describe morphologically similar microorganisms.

Results

The investigations revealed new, previously undiscovered sources of contamination in all areas. The bacterial contamination of the production and water pipes was significantly reduced by means of UV irradiation. The qualitative description showed unexpectedly complex bacterial contamination in all sample groups.

Conclusion

It is reasonable to assume that the methods established in the food industry are suitable in the paints and coatings and cosmetics sectors. Further investigations could identify weaknesses in the methods and help to develop suitable combinations of different methods that can be used as sustainable preservation methods.

Comparative fermentation of seven non-dairy milk alternatives using an in-house developed vegan starter culture

Dipl. Ing. ETH Helena Stoffers¹, Gabriela Purtschert², Ghazal Nemati³, Barbara Guggenbühl⁴, Barbara Walther⁴, Devon Jakob², Ueli von Ah²

¹Applied processing technology, Agroscope, ²Biotechnology, ³risk assessment and mitigation,

⁴Human nutrition, sensory analysis and flavour

Objective:

As non-dairy alternatives gain popularity, nevertheless they continue to face challenges such as nutritional imbalances and flavor limitations. Fermentation can improve sensory profiles, nutritional value, microbial safety, bio-accessibility of protein and synthesizing vitamins. However, existing starter cultures for dairy products might not be appropriate, necessitating further studies. This study aimed to compare the fermentation of seven non-dairy alternatives using an in-house developed vegan starter culture. We explored the influence of carbohydrates and proteins on fermentation profiles.

Methods:

We monitored pH at five-minute intervals for 18 hours at 30°C using established fermentation protocols. The substrates included potato, pea, oat-pea, oat, hazelnut, soya, and lupin drinks, with standard organic low-fat milk as a control. The nutrient content was taken from the product labels. The vegan starter mixture comprised *Lactobacillus delbrueckii subsp. lactis*, *Streptococcus thermophilus*, *Lactococcus spp.*, and *Leuconostoc spp.* Selected sensory properties of the samples were evaluated quantitatively and qualitatively before and after fermentation.

Results:

Rapid fermentation was observed in oat, oat-pea based, and hazelnut-based drinks due to their higher sugar content, leading to faster fermentation rates compared to the other plant drinks. In contrast, potato and lupin-based drinks showed a prolonged adaptation phase. For potato-based drink likely because the high initial pH inhibited the growth of *Lactobacillus delbrueckii subsp. lactis* and *Lactococcus spp.*, and slowed down *S. thermophilus*. Pea drink had a slow fermentation with minimal pH change over 18 hours, due to its low carbohydrate content (0.3 g per 100 g). Soy-based drinks and the organic milk displayed similar fermentation patterns. After 18 hours, oat and hazelnut drinks showed a lower pH compared to organic milk, influenced by their low protein content, which reduces buffering capacity and thus, results in a lower final pH. Meanwhile, pea and oat-pea drinks, with higher protein content, recorded the highest final pH values.

Conclusion:

Plant-based drinks differed from dairy and within each other due to various characteristics such as buffering capacities, initial pH, carbohydrate and protein content as well as sensory characteristics. This necessitates tailored fermentation criteria and bacterial selection in order to produce plant-based fermented products which are accepted by consumers.

Effect of varied *Kluyveromyces marxianus*-to-*Saccharomyces cerevisiae* co-inoculum ratio on the aroma composition of Gewürztraminer wines

Nicola Cappello¹, Riccardo Savastano^{2,3}, Mauro Paolini¹, Filippo Amato^{3,4}, Eric Fouquet², Nicolas Vivas⁵, Nicola Francesca³, Silvia Schiavon¹, Raffaele Guzzon¹, Roberto Larcher¹, Tomas Roman¹

¹Fondazione Edmund Mach - Center for Technology Transfer, ²Université de Bordeaux - Institut des Sciences Moléculaires, ³Università degli Studi di Palermo - Department of Agricultural, Food and Forest Sciences, ⁴HTS Enologia, ⁵Université de Bordeaux - Centre de Recherche Demptos

Aim:

The past decades have witnessed a heightened focus on non-*Saccharomyces* yeasts in the winemaking and biotech industries. This is attributable to the presence of certain highly or under-expressed distinct features that can be leveraged in the winemaking process. Upon selection, the suitability of a strain in industrial context is not solely determined by its attributes, but rather by its overall performance while meeting product expectations. The work studied the effect of the *K. marxianus*-to-*S. cerevisiae* inoculum ratio of must on the volatile composition of the resulting wines.

Method:

A Gewürztraminer grape must was inoculated at $2 \cdot 10^6$ CFU/mL with five different ratios of the Km L2009 *K. marxianus* strain (n=3): 80%; 90%; 95%, 99% and 99.9%. Results were also compared to those obtained from the pure *S. cerevisiae* fermentation and the sequential protocol with *S. cerevisiae* three days after *K. marxianus* was inoculated. The volatile composition of wines were analysed by GC-MS/MS at the end of fermentation.

Results:

In comparison with *S. cerevisiae*, *K. marxianus* wines exhibited higher concentrations of short and medium chain fatty acids and their corresponding ethyl esters, while acetate esters were lowered. Between inoculation protocols differences emerged for acetate esters (80% wines were richer than 95% and 99.9%) and fatty acid (99.9% was richer than 80%). Moreover, the sum of acetate esters showed a downward trend with the increasing *K. marxianus* inoculum ratio while the sum of fatty acids an upward one ($p < 0.05$), even if ethyl esters pattern was not discernible in relation to the number of *K. marxianus* viable cells inoculated. In relation to grape-derived volatiles, neither norisoprenoids nor terpenes were distinguished between inoculation protocols; however, for the latter, the primary aroma compounds indicative of Gewürztraminer's aroma tipicity, it was displayed a similar positive trend ($p < 0.05$) with the increased ratio. The first component of the Principal Component Analysis applied to the overall dataset clearly differentiated wines on the basis of the presence of *K. marxianus*, while the second one distinguished the inoculum ratio.

Conclusion:

The *K. marxianus*-to-*S. cerevisiae* co-inoculum ratio emerges as a novel strategy to modulate Gewürztraminer aromatic profile.

Sesame Processing Refuse: From waste to functionality

Professor Mohamad Abiad¹, Reem Farhat

¹American University Of Beirut

Aim: As people become more conscious of the environmental impact of their food choices, many are shifting towards plant-based diets. While acceptable substitutes have been created for non-fermented dairy products, there's still a gap in satisfying alternatives for fermented dairy like yogurt, aged cheeses, and semi-soft cheeses. Extensive research has been devoted to exploring the remarkable functional attributes of proteins. To examine how sesame protein isolate affects dairy products, we investigated the functional characteristics of sesame protein isolate, including its emulsifying, foaming, and antioxidant properties. The current work focuses on recycling sesame processing waste through protein extraction and modifying its functionality to offer distinctive properties within the hybrid cheese structure and to contribute to the overall sensory attributes and shelf-life stability.

Method: Following protein extraction from sesame processing waste, it was incorporated into soft cheese formulations, and sensory qualities and shelf-life stability were assessed.

Conclusion: Investigating the functional properties of sesame protein isolate (SPI) holds promise for enhancing soft cheese production. Incorporating SPI into soft cheese formulations can contribute to soft cheeses' sensory attributes and shelf-life stability. This research underscores the importance of exploring alternative protein sources to meet consumer demand for high-quality and nutritious dairy products.

Breathing the gap between meat by-products and sustainability

Dr Sara Baptista-silva¹, Maria Martingo¹, Inês Cruz², Manuela Pintado¹

¹Universidade Católica Portuguesa, CBQF - Centro de Biotecnologia e Química Fina – Escola Superior Biotecnologia, ²Grupo Primor / Primor Charcutaria-Prima

Aim: Over the past 20 years, global meat consumption has increased steadily by 58%, which means that approximately 18 million tons of meat by-products are produced and are costly to handle in an eco-friendly way. Swine kidneys are notable by-products within the meat industry, renowned for their nutrient-rich composition comprising proteins, vitamins, and minerals, making them a potential ingredient in the food sector. However, the efficacy of their use is frequently impeded by the ammonia content, a hazardous by-product stemming from post-slaughter protein metabolism accumulation. Elevated ammonia concentrations compromise its sensory characteristics and pose notable public health risks.

Method: For ammonia reduction, six pre-treatment methods were studied, regarding the use of hydrogen peroxide, acetic acid and sodium bicarbonate. Concentrations of 1-12% (w/v) with different neutralization and washing procedures between 30 min, 1, 2, 3 and 24 h were tested for the above solutions. Ammonia detection was performed using an ammonium ion selective sensor, and a magnesium sulfate calibration curve ranging from 0.0312 to 1 g/L. Also, a prototype mortadella product was developed using 20% kidney mass to test the method's effectiveness and safety.

Results: The best results of ammonia reduction were achieved for sodium carbonate solutions, varying between 20 and 65%. The greatest reduction in ammonia content was 65%, using a sodium carbonate solution, in a bath for 2 h. The application of treated kidneys in a prototype of mortadella product led to a reduction in the ammonia content of 70%, which represents ca. 2 mM in the final product.

Conclusion: This study developed a new methodology for swine kidney pre-treatment, as a meat industry by-product, promoting its safe consumption, with improved odour and flavour.

This research underscores the importance of recognizing the value of by-products derived from the meat industry as a mean to further sustainability goals and develop innovative food products with significant added value, addressing both nutritional and economic considerations.

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Enzymatic Processing of Hazelnut Skins by treatment with carbohydrases

Dr Ester De Martino¹, Dr Angela Sorrentino², Professor Prospero Di Pierro^{1,2}

¹Department Of Agricultural Sciences, University of Naples "Federico II", ²Caisial (Centro di Ateneo per l'Innovazione e lo Sviluppo dell'Industria Alimentare)

Aim:

Italy is an important producer of hazelnuts and ranks second in the world after Türkiye. This results in the production of large quantities of waste with a bad impact both on economic and ecological aspects. However, among residues and wastes hazelnut skins represent a good source of fibres that still may be utilized to recover molecules of interest and put them in new formulation. From preliminary characterization, the carbohydrate fraction in hazelnut skins resulted about 70% of the material. Since one of the greenest and most innovative strategy to break down chemical bonds and hydrolyse molecules are Enzymatic biotechnologies, carbo-hydrolases were chosen for enzyme processing of hazelnut skins.

Method:

Micronized hazelnut skins were subjected to enzymatic treatment with pectinase, xylanase, cellulase and hemicellulase. The assay incubation was performed at different conditions of pH, time, temperature and solid:liquid *ratio*. The effect of enzyme hydrolysis was assessed both on liquid and solid fractions by evaluating released sugars (by DNS and Dubois analysis) and changes in the morphological structure of the remaining solid material (by SEM and ATR-FTIR).

Results:

Results show that all enzymes allowed the release of reducing and total sugars from the raw matrix, although xylanase and hemicellulase were more efficient. SEM images of treated samples revealed that the surface of the materials resulted smoother compared to the controls. ATR-FTIR investigations are ongoing but preliminary results highlight differences in the spectra of samples subjected to enzymatic digestion compared to controls.

Conclusion:

In a circular economy design, the proposed enzymatic treatments represent a good strategy for the valorisation of waste and residues from hazelnut processing. In the next step, residual material after enzymatic digestion will be characterized for the hydration and water solubility indices. Also, the presence of phenolic compounds, as well as their antioxidant capacity against DPPH, will be assessed on the liquid fractions. This approach leads to the recovery of fibre and/or bioactive compounds useful in the preparation of functional foods.

New approach to apple pomace byproduct: developing fermentable must for NoLo beverages.

PhD Shuyana Deba¹, Unai Aguirre-Cano¹, Aitor García-Roldán¹, Iratxe Olazaran-de la Peña¹

¹Food Technology Department, Leartiker S. Coop.

Aim:

The cider industry is one of the leading producers of apple pomace byproduct in the Basque Country, which makes up a significant portion of food waste. However, it contains polyphenols, simple and complex carbohydrates, pectin, significant amounts of major and trace minerals, and fats, making it suitable for fermentation. The aim was to characterize the must obtained from this byproduct for the development of a non- or low alcohol (NoLo) fermented beverage, offering a new product idea for local cideries.

Method:

Apple pomace was purchased from local cideries after the pressing process and preserved frozen. The must was obtained by maceration at a ratio of 1:2 (water:apple pomace) at 10 °C and the effect of Rapidase PL Smart pectinase was also studied. Samples at 2, 4, 6 and 8 hours were characterized: total sugar, malic acid, pH, density, total phenols, and total acidity. Then, fermentation was carried out with co-inoculation of *L. plantarum* and *Pichia kluyveri*, and water kefir grains for final product design and preliminary sensory analysis were done.

Results:

No significant effect was observed in sugar and other compounds release with different maceration times and pectinase use. The must is composed mainly by sugar (around 21 g/L) and malic acid (1.1 g/L) as fermentative substrates, polyphenols (0.18 g/L) and aromatic compounds that provide the must with apple flavour. Fermentation trials with kefir grains resulted on the reduction of sugars to 8.4 g/L and the production of CO₂, suggesting that alcoholic fermentation occurred. Instead, the co-inoculated fermentation trials did not consume sugar neither produce CO₂ and a reduction of malic acid was produced, reaching levels of 0.05 g/L. Both beverages were tasted and were positively evaluated.

Conclusion:

By macerating apple pomace for 2 hours at 10°C a fermentable must is obtained, which can be used to develop low-alcohol beverages by alcoholic and malolactic fermentations. Therefore, this study shows the potential of fermentative processes to obtain new fermented beverage revalorizing food byproducts, as apple pomace, and opening innovative products in the market.

Drying process impact on the functional properties of unfractionated potato peels: a comparative study

Emma Dieterlen¹, Nawal Dagher², Adrien Izzet¹, Marine Masson², Delphine Huc-Mathis²

¹Inrae, ²AgroParisTech

Aim:

Potato (*Solanum tuberosum* L.) peel is a byproduct generated by the peeling of raw tuber, a common process in the industry of potatoes. It represents a great amount of waste usually discarded (Sampaio et al., 2020) although a concentrated source of nutrients such as starch, dietary fibres and proteins (Joshi et al., 2020). In order to minimize the environmental impact, what about making a circular use of this byproduct as clean-label ingredient for food applications? The aim of this study is to compare the physicochemical characteristics of two powders that were obtained with different drying time and temperature combinations and to explore various applications.

Method:

Both powders were obtained only by drying and grinding. One half of potato peels were dried at 180°C for 30 min (PPP180) and the other half at 50°C for 5h (PPP50) to mimic aggressive and slow drying respectively. Potato starch was used as a reference to evaluate the difference between a cracked product and a heterogeneous byproduct with a complex composition. To characterize the powders, the pH, Brix degree, soluble content and the water holding capacity were measured. Powder colour was measured with ImageJ. Meanwhile, the powders were used in food applications as a substitute for potato starch (a thickener, texturizer in patisserie and noodles) and as a functional ingredient (texturizing and nutritional additive). The applications were voluntary very diverse.

Results:

The physicochemical characteristics are significantly different between powders and starch however only the pH of the supernatant significantly differentiates the two PPP powders with an acidic pH at 5.45 for PPP50 and at 6.18 for PPP180. To the senses, the two PPPs are differentiated by their smell and colour due to the Maillard reaction which makes applications with PPP180 sensory blocking such as chocolate mousse, mayonnaise and noodles. When it's heated, PPP50 does display thickening power equivalent to that of starch, while it is less concentrated in starch. It is an interesting substitute for starch, but the food recipes need to be improved using sensory engineering.

Conclusion:

These promising results make it possible to establish decision support when drafting a protocol for the valorisation of potato peels.

Adding value to Capsicum pepper byproducts as natural antimicrobial for product development

Marta Silva⁴, Sofia Fonseca⁴, Norton Komora³, **Patrícia Fradinho**^{1,2}, Andreia Cavaco¹, Catarina Prista^{2,4}

¹Colab4Food - Collaborative Laboratory for Innovation in the Agri-Food sector, Rua dos Lagidos, 4485-655 Vairão, ²LEAF—Linking Landscape, Environment, Agriculture and Food Research Center, Associate Laboratory TERRA, Instituto Superior de Agronomia, Universidade de Lisboa, 1349-017,

³Casa Mendes Gonçalves, Zona Industrial, Lote 6, 2150-268, ⁴Instituto Superior de Agronomia, Universidade de Lisboa

Aim:

The industry processing of hot peppers aims to produce a pepper fermented paste for application in sauces and condiments. This process generates around 20% of waste (seeds, skin, and pulp) which are currently discarded contributing to energy and water losses.

Several studies describe capsaicinoids as the responsible biomolecules for the pungency and antimicrobial properties of Capsicum peppers. This study assesses the antimicrobial activity of several Capsicum pepper byproducts against different food spoilage microorganisms to give these wastes a second life by using them as natural preservatives.

Method:

To test the ability of different yeasts, bacteria, and fungi to grow on byproduct of different peppers and to test their antimicrobial activity, pepper byproducts (CR, RH, OH, YH, WH, JP varieties) were milled and solidified with agar with and without the proper culture medium to create pepper-base media with different concentrations (10%, 25%, and 50%). Drop test assays were performed using 10-fold diluted suspensions up to 10^{-6} and spotting 3 μ L of each suspension with a replica platter on the prepared plates. Plates were incubated at 30 and 37°C, and growth was monitored after 3 days, 1 and 2 weeks.

The most promising byproduct pepper varieties in terms of antimicrobial properties were used in the development of spicy mayonnaises without artificial preservatives and compared with the company's current commercial formula in terms of the microbiological stability.

Results:

The preliminary results of the antimicrobial assays demonstrated that all pepper varieties tested had an inhibitory activity on all the bacteria tested, although they do not affect the growth of these microorganisms at the same level. Regarding yeast inhibition, CR, OH and JP varieties seem to have antimicrobial effect at all concentrations tested. In addition, the highest pepper concentration of all Capsicum varieties inhibited the fungi tested.

Conclusion:

The pepper byproducts presents antimicrobial properties against several food spoilage microorganisms, and could be valued for innovative food development and as natural preservatives while contributing to the waste reduction in the agribusiness.

Acknowledgements:

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Re-valorization of grapeseed press cake for food applications

Dr. Maria Gräfenhahn¹

¹Institute of Life Technologies, University of Applied Sciences and Arts Western Switzerland Valais-Wallis (HES-SO VS)

Aim:

Grapes, particularly the species *Vitis vinifera* L., are one of the most widely grown crops in the world and are mainly used for the wine industry. Grape pomace is its by-product and contains > 30% grape seeds. Grape seeds are mainly composed of lipids approx. 22% and carbohydrates and proteins approx. 12%, respectively (in the DM). More recently, this by-product of wineries has been used for the extraction of the seed oil and natural antioxidants. However, this is only a small fraction of it and most of the grape seeds remain unutilized. After the oil extraction process a defatted grapeseed flour is obtained containing approx. 20% carbohydrates, 11% protein, and over 50% fiber. Even though this product is, in some regions, available for consumers, it is not part of many if any, commercially available food products. A reason for this might be, the composition including protein and fibers and their interactions, which influence the functionality.

Approach:

One approach to increase the application field of this material would be to separate them into single components, i.e., proteins and fibers. Few publications focusing on the extraction of proteins from *Vitis vinifera* L. have shown that the amino acid composition and properties are very similar to those of cereals and other oilseeds. Thus, proteins extracted thereof could be used as a substitute in protein-rich foods, e.g., meat or dairy analogues.

Results:

In this contribution, we will present the results of the physicochemical and functional characterization of grapeseed press cake from two grape varieties grown in Valais. The results show that both the protein- and fiber-rich extracts have good functional properties, including solubility, interfacial, and thickening properties compared to other plant proteins.

Conclusion:

These results will be presented in detail and the implications for the use of the grapeseed proteins in foods will be discussed.

Effect of pulsed electric fields on molasses yield from orange peel waste

Sofia Chanioti¹, Dr. Varvara Andreou¹, Panagiotis-Konstantinos Masouras¹, Marianna Giannoglou¹

¹Institute Of Technology Of Agricultural Products Elgo-demeter

Aim:

The production of molasses from orange peels waste involves the press liquor obtained during the pressing process, which is the liquid extracted from orange peels waste before their drying. Instead of discarding this valuable liquid, it undergoes further processing to recover components such as sugars. This liquid is evaporated, concentrating the sugars present and resulting in the formation of molasses. Molasses is rich in sugars and other compounds extracted from the orange peels, suitable for various applications such as a feed additive. However, evaporation is a time-consuming step with low yields. Pulsed Electric Fields (PEF) have the potential to enhance the extraction of intracellular compounds, including residual sugars, from orange peels by permeabilizing their cells. This process can result in the production of press liquor with higher sugar content, consequently increasing the yield of produced molasses.

The objective of this work was to evaluate the potential benefit of PEF as pretreatment for pressing orange peels waste, targeting to increased yield of produced molasses and reduction of the evaporation time, resulting in energy savings.

Method:

Orange peels waste was subjected to selected PEF conditions (2.5 kV/cm, 1000 pulses, 100 Hz, 15 μ s) as pretreatment. PEF treated and untreated orange peels waste were immersed in a bath solution of 0.1% CaCl₂ for 30 min. Pressing process of orange peels was followed to extract liquid rich in sugars. The obtained liquids were then evaporated to achieve a concentration of 62 °Brix. The yield of molasses was determined for both samples. All samples were analyzed for their individual sugars and organic acids, as well as their bioactive compounds.

Results:

The liquid obtained from PEF treated orange peels had significantly higher initial total soluble solids (6.0 °Brix) compared to the untreated one (5.2 °Brix). This indicates that PEF pretreatment led to increased yield of molasses, from 6.8% to 8.2% and reduced the evaporation time to reach the desired °Brix level of molasses.

Conclusions:

The results obtained in this study demonstrate the potential of PEF pretreatment to increase the yield of produced molasses while simultaneously accelerating the evaporation process, thereby minimizing energy consumption.

Bio-accessibility and stability study of nanoencapsulated phenolics and carotenoids from olive and tomato pomace

Dr Maria Katsouli¹, Ioanna Thanou¹, Dr Athina Ntzimani¹, Eugenia Raftopoulou¹, Prof. Maria Giannakourou¹, **Professor Petros Taoukis¹**

¹National Technical University of Athens

Aim:

Valorization of agricultural by-products via extraction of bioactive compounds presents a sustainable approach to enhance food functionality and mitigate waste. Agricultural by-products, such as olive and tomato pomace, are abundant sources of valuable compounds, including phenolic compounds and lycopene. Extraction of these compounds not only adds value to what would otherwise be considered as waste, but also contributes to the development of novel foods with enhanced nutritional profiles and potential health benefits. This study investigates the encapsulation of bioactive compounds extracted from olive and tomato processing by-products via oil-in-water (O/W) nanoemulsion formulations. The aim is to enhance the stability and bioaccessibility of these valuable compounds for potential applications in functional foods and nutraceuticals.

Method:

O/W nanoemulsions consisted of an aqueous phase: 8% wt. Tween 80, 1% wt ascorbic acid and 1% wt phenolic compounds extracted from olive pomace, and a lipid phase: 10% wt. pomace oil enriched with 1% wt lycopene extracted from tomato by-products. Two steps of homogenization process were used for the O/W nanoemulsions, employing high-speed and high pressure homogenization techniques. Stability studies were carried out at 25 and 4 °C. Additionally, the pH values of nanoemulsions were adjusted at 2-7, using HCl or NaOH solution. Physicochemical properties (droplet size), encapsulation stability (ES), and bioaccessibility (INFOGEST protocol) of nanoemulsions were evaluated during storage.

Results:

O/W nanoemulsions enriched with phenolic compounds and lycopene remained stable and in nanorange (mean droplet diameter ca. 250 nm), indicating the robustness of the encapsulation system in maintaining the concentration of lycopene (>70% ES) over time. Bioaccessibility assessment demonstrated enhanced release of phenolic compounds and lycopene from the nanoemulsions, suggesting improved bioavailability compared to non-encapsulated forms. Storage conditions (pH, temperature) influenced the physicochemical properties and ES of the nanoemulsions. Those adjusted at pH 5 exhibited optimal ES for both phenolic compounds and lycopene and stable droplet size, highlighting the importance of pH optimization in nanoemulsion-based delivery systems.

Conclusion

Oil in water nanoemulsions are deemed effective carriers for bioactive compounds from agricultural by-products, offering enhanced stability, encapsulation efficiency, and improved bioaccessibility. This research contributes to the development of sustainable strategies for valorizing food waste and improving the functional properties of food products.

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A traditional fermented dry-cured sausage with a bioactive ingredient from wine pomace

PhD María Jesús Martín Mateos¹, Phd Matilde H. D'Harrigo Huapaya¹, Miriam Sánchez Ordóñez¹, Phd Jonathan Delgado Adámez¹, Phd Jesús Javier García Parra¹, PhD María Rosario Ramírez Bernabé¹
¹Cicytex

Aim:

The “salchichón” is a traditional fermented dry-cured sausage which is made by mixing minced meat with seasonings and undergoes a prolonged drying maturation process before consumption. It's a common industrial practice to also include some synthetic additives, such as nitrites, to improve microbial stability and safety, color, and flavor. However, consumers are demanding more natural and additive-free meat products. Grape pomace is the main by-product of winemaking, and it is rich in phenolic compounds with antioxidant and antimicrobial activity. Due to its properties, it could reduce the need of adding synthetic additives in meat products. Therefore, the main objective of this study is to evaluate the incorporation of grape pomace into dry-cured sausages to improve their preservation and replace the use of synthetic additives like nitrifying salts.

Method:

A bioactive ingredient was obtained from valorised red grape pomace (cv *Tempranillo*) through thermal blanching and hydrostatic high-pressure treatment. This ingredient (RGP) was added in the “salchichón” formulation. Four formulations were evaluated: Negative Control (NC: without RGP or synthetic additives), Positive Control (PC: with ascorbic acid and nitrites), Low Level (LL: 0.5 %) and High Level (HL: 1%) of RGP. Microbiological counts, instrumental colour, lipid and protein oxidation were analysed. The effect of RGP on sensory characteristics was analysed.

Results:

The ingredient obtained was rich in fibre and phenolic compounds. In “salchichón”, the RGP favored the growth of lactic acid bacteria and presented some antioxidant properties. It reduced the oxidation of the sausages, but the red color of those manufactured with nitrifying salts was not reached. Tasters no detected sensory differences (taste, flavor) with the inclusion of RGP.

Conclusion:

The inclusion of RGP in the formulation of the “salchichon” favored an adequate fermentation similar to that of sausages that included nitrites and ascorbic acid. In addition, RGP ingredient presented a slight lipid antioxidant effect on the dry-cured sausages. The addition of the ingredient did not negatively affect the sensory perception of the sausage, however, the color of those manufactured with nitrifying salts was not reached.

Valorisation of various cassava processing residues to produce value-added bioproducts

Ms. Andreia Massamby^{1,2,3}, Volkmar Passoth¹, Bettina Müller¹, Johanna Blomqvist¹, Su-lin Leong¹, Tivana Lucas^{2,3}, Macuamule Custódia², Mats Sandgren¹

¹Swedish University Of Agricultural Sciences, ²Eduardo Mondlane University, ³Centre of Excellence in Agri-Food Systems and Nutrition, Eduardo Mondlane University

Aim:

Cassava is a starch crop largely produced and consumed in Africa. In Mozambique, this crop is ranked as the most important staple food making up almost a third of calories consumed after maize. In Inhambane province, cassava processing activities take place in several cassava farmers associations and at the main cassava starch mobile factory, generating large amounts of residues, both in solid and liquid forms. The liquid waste (cyanide-containing cassava press water) and solid waste (cassava peels and cassava fibers) are indiscriminately discarded without any treatment rather than being used to produce high value-added bioproducts.

This study aimed to evaluate the efficiency of converting cassava residues in two types of hydrolysates (cassava peel hydrolysate and cassava fibers hydrolysate) through enzymatical hydrolysis into substrates for feed and food applications.

Method:

Two oleaginous yeast strains, *Saccharomyces cerevisiae* and *Rhodotorula toruloides* were tested as potential microbes to convert the available sugars in cassava hydrolysates to value-added bioproducts such as ethanol, lipids, and proteins. FTNIR was used to determine intracellular lipid production in yeasts, and HPLC to determine the concentration of reducing sugars and ethanol.

Results:

Keeping the pH stable at 6.0 and adding Ammonium sulphate in treatments with peel and fiber hydrolysates was crucial for maximising growth of the tested strains. *S.cerevisiae* showed growth activity even in treatments with pure cassava peel hydrolysate whereas *R.toruloides* could grow in treatments with pure fiber hydrolysate.

Conclusion:

Our results aim to validate sustainable ways to reutilize these residuals, giving local farmers better solutions to minimize the impact of this waste on soil, environment, and human health, increasing the local productivity of cassava.

Valorization of protein-rich food processing sidestreams: spectroscopy-based methodologies for collagen content determination

Greta Nardini¹, Kristian Hovde Liland¹, Sileshi Gizachew Wubshet², Nils Kristian Afseth², Kenneth Aase Kristoffersen¹

¹NMBU - Norwegian University of Life Sciences, ²NOFIMA - Norwegian Institute of Food, Fisheries and Aquaculture

Aim:

Despite alternative protein sources, the global consumption of meat proteins is projected to increase by 14% by 2030. Poultry meat represents the primary driver and is expected to account for 52% of the additional meat consumed. Because of the primary poultry contribution to the growth production, the greenhouse gas emissions of the meat sector are calculated to increase by only 5% by 2030. The shift toward poultry reflects the significant roles it plays in several populous developing countries that choose it for the low price, but in higher-income countries it is favored because of its higher protein/lower fat content.^[1]

The Sustainable Development Goals of the United Nations include: “2. Zero Hunger” in which targets 2.1 and 2.4 emphasize sustainable food production that increases productivity and production ensuring all people access to nutritious and sufficient food; and “12. Sustainable consumption and production patterns”. Thus, poultry plays a crucial role in addressing food security and nutrition challenges, but to achieve these goals the food industry must implement new methods and technologies to reduce waste and improve the valorization of its sidestreams.

Method:

Enzymatic Protein Hydrolysis (EPH) is a well-established and versatile technology for improved utilization and valorization of high-quality protein from food industry sidestreams. Low-value cuts from poultry processing, a protein-rich biomass, are transformed into complex protein hydrolysates with different physicochemical properties through the EPH process. For example, collagen-enriched protein hydrolysates have been widely utilized for food products, pharmaceuticals, medical products, and cosmetics.^[2]

To obtain the desired product with specific quality and optimal yield, it is vital to apply fast non-destructive methodologies for monitoring and adjusting the process.^[3]

Results:

In this study, Nuclear Magnetic Resonance (NMR) spectroscopy has been used to gain a deeper understanding of the EPH process of poultry sidestreams by following the solubilization of collagen.

Conclusion:

NMR spectroscopy has been used here for investigating collagen content. It is a powerful technique that provides both qualitative and quantitative insight into the composition of complex matrices. This approach is expected to be an important and comprehensive tool in the growing industry segment of sustainable utilization of protein-rich biomass.

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The Use of Poppy Seed Cake in the Preparation of Functional Products

Nujamee Ngasakul¹, Ali Kozlu¹, Diana Karina Baigts Allende¹, Sandra Teresita Martín del Campo Barba¹, Iveta Klojdová¹

¹Czech University of Life Sciences Prague

Aim: Poppy seed cake, a valuable by-product from oil extraction, is rich in fiber, protein, and phenolic compounds; it contains some vitamins, fat, microelements, and macroelements. Due to the significance of these constituents, this investigation aims to valorize this by-product by recovering its bioactive compounds, characterizing them, and incorporating them into functional products, namely model desserts based on the emulsion.

Method: The poppy seed cake was pre-treatment using a hot-air oven (130°C for 25 min) and microwave (600W for 15 min). Subsequently, bioactive compounds were extracted from the poppy cake using two different concentrations of ethanol, employing ultrasound-assisted extraction at 40°C for 1 h in an ultrasonic bath. The extracts were then analyzed for their phytochemical composition and in vitro antioxidant activity. A functional model product based on emulsion was developed incorporating the selected extract, followed by an evaluation of the stability and activity of the bioactive compounds.

Results: The extract obtained from absolute ethanol exhibited a colorless transparent appearance compared to that obtained from 50%v/v ethanol and distilled water. Analysis of the extracts revealed the presence of phytochemicals such as phenolic acids and flavonoids. Moreover, all samples demonstrated antioxidant activities, including free radical scavenging activities (DPPH and ABTS), ferric reducing power (FRAP), and metal chelating activity (MCA). The developed model functional products based on emulsion demonstrated the bioactivity as well.

Conclusion: These findings highlight the potential of extracts derived from poppy seed cake as a promising source of bioactive compounds, which could be expected for the development of functional products with bioactive properties.

Development of a chitosan active biopolymer with thymus essential oil

Dr Rosario Ramirez¹, Irene Orts¹, Bruno Navajas¹, Javier Rocha¹, Sara Martillanes¹, María Cabeza de Vaca¹, JONATHAN DELGADO-ADÁMEZ¹

¹Cicytex (centro De Investigaciones Científicas Y Tecnológicas De Extremadura)

Aim:

Active packaging represents an innovative strategy aimed at extending the shelf life of food products. Chitosan is an eco-friendly packaging material derived from the valorization of waste from the agri-food industry. It is a cationic polysaccharide soluble in acidic solutions, possessing film-forming capacity and intrinsic antimicrobial activity due to its chemical structure. Chitosan is a highly versatile polymer, whose characteristics and properties have made it the subject of study in the development of active materials. On the other hand, essential oils comprise a blend of natural, volatile, and fragrant compounds extracted from various plants. They possess antimicrobial, antioxidant, anti-inflammatory, and immunomodulatory properties. The genus *Thymus* L., renowned for its aromatic qualities, is abundant in the Mediterranean region. The main objective of this study is to evaluate the properties of essential oil derived from *Thymus vulgaris* and to assess the antimicrobial and antioxidant efficacy of a chitosan biopolymer with this essential oil.

Method:

Thymus vulgaris essential oil was extracted through hydrodistillation or steam distillation. The composition of the essential oil was characterized by SPME-GC-MS and its antimicrobial properties against *Escherichia coli* and *Listeria innocua* as well as its antioxidant activity were evaluated. Chitosan films were prepared dissolving chitosan (2% w/v) with 1% of 80% lactic acid. Subsequently, glycerol was added to the solution at 50% (w/w). Finally, different concentrations of the essential oil at 0- 3%, were added to the mixture. The resulting solutions were poured into Petri dishes and dried until film formation. The antioxidant activity of the developed biopolymers was evaluated by the ABTS method.

Results:

A 97% of the essential oil could be characterized by SPME-GC-MS and the most abundant compounds identified were carvacrol, p-cymene and thymol. The essential oil presented high antimicrobial activity against *E. coli* and *Listeria innocua* even at dilutions 1/100. *Thymus* essential oil presented high antioxidant activity. The essential oil was included in a chitosan biopolymer which retained the antioxidant activity even at the lowest doses of essential oils.

Conclusion:

Thymus essential oil presented great antioxidant and antimicrobial properties. The addition of essential oil to chitosan biopolymer provided antioxidant activity to films. Therefore the developed chitosan biopolymer with *Thymus* essential oil could be utilized to increase the shelf-life of foods like meat products, which are highly perishable, due to the development of microorganisms and lipid oxidation reactions.

formation of polycyclic aromatic hydrocarbons in burgers with a white grape pomace ingredient

Dr Rosario Ramirez¹, Juan Ramón Maroto¹, Matilde D'Arrigo¹, Miriam Sánchez-Ordóñez¹, María Díaz¹, Beatriz Toro¹, M. Jesús Martín-Mateos¹

¹Cicytex (centro De Investigaciones Científicas Y Tecnológicas De Extremadura)

Aim:

The polycyclic aromatic hydrocarbons (PAHs) are a group of organic compounds containing two or more fused aromatic rings which are genotoxic, mutagenic and carcinogenic. The occurrence of PAHs in meat is associated with thermal treatments, from the smoke generated by the incomplete combustion of fuels (such as wood or charcoal) and deposited on the surface of the meat. The composition of the meat products also plays an important role. Some studies have associated meat products with more antioxidants to low contents of PAHs. Natural products containing antioxidants like polyphenols could inhibit the production of PAHs. Winemaking by-products present high levels of polyphenols, which could be successfully preserved by non-thermal technologies like hydrostatic high pressure. The main advantage of the use of this technology is that the process allows an integral use of the pomace (the whole by-product) so that no residues are generated. The main objective of the study was to evaluate the effect of the inclusion of an ingredient from white wine pomace (WWP) in the formation of PAH in barbecued pork burgers.

Method:

White wine pomace (cv *Cayetana*) was valorized by hydrostatic high pressure in order to obtain a bioactive ingredient. This ingredient from WWP was added to pork burgers in order to improve their preservation. Five formulations of burgers were manufactured: Control (burgers without metabisulphite); commercial (burgers manufactured with metabisulphite); burgers manufactured with 0.5% (w/w) grape pomace; burgers manufactured with 1% (w/w) wine pomace; and burgers manufactured with 3% (w/w) wine pomace. They were cooked with charcoal briquettes in barbecue. Proximate composition and bioactive compounds content of the WWP ingredient was analysed. Lipid oxidation (TBA-RS) and the levels of PAH (HPLC-DAD-FLD chromatography) in burgers were analysed.

Results:

The ingredient from WWP presented high phenolic compounds content and antioxidant activity. Barbecued burgers with the developed ingredient presented lower TBA-RS values than control and burgers manufactured with metabisulfite. In addition, they had also the lowest values of PAHs, like phenanthrene and fluorene.

Conclusion:

The manufacture of pork burgers with a valorized bioactive ingredient from white wine pomace could prevent lipid oxidation and the formation of PAH during barbecue.

Red pepper and plum valorization by hydrostatic high pressure for preservation of fresh meat products

Miriam Sánchez Ordóñez¹, María Cabeza de Vaca¹, Jesús Javier García Parra¹, María Díaz Ponce¹, Jonathan Delgado Adámez¹, Rosario Ramírez Bernabé¹

¹Centro De Investigaciones Científicas y Tecnológicas de Extremadura

Aim:

Pepper and plum foods are rich in phenolics and antioxidants compounds that reduce oxidative stress and protect against chronic diseases such as cancer and cardiovascular disease. By-products generated in industry can be utilised and valorised, producing a circular economy. High Hydrostatic Pressure (HHP) is a clean technique that maximises product utilisation and preserves bioactive compounds. The preservation of meat products needs to include additives such as sulphites and nitrites to prevent spoilage. However, consumers demand more natural and healthier products. The aim is to solve the demands of the meat industry that need to incorporate ingredients for the preservation of their products replacing chemical additives with antioxidant and antimicrobial natural ingredients.

Method:

The by-products of plum (var. Crimson Globe) and red pepper (var. Franchi) were used in the form of puree. First, the plum was scalded (80°C for 1 min) to inactivate polyphenol oxidase (PPO). No PPO activity was found in the pepper analyses. Subsequently, HHP treatment (600 MPa 5 min) was applied to the pepper puree and blanched plum puree to obtain a revalorised ingredient. The burgers were made with minced pork and spiced. Following this, a group with sulphites (with sulphites (0.45 g kg⁻¹)) was prepared and subsequently groups with different levels (1% and 5%) of the plum and pepper ingredients were prepared. Six lots were analysed: control, sulphites, pepper L (1%), pepper H (5%), plum L (1%) and plum H (5%). Chemical composition of the ingredients were studied. Phenolic compounds and lipid oxidation of burgers with different formulations was analysed.

Results:

The ingredients showed significant fibre content, acidity, moisture and high content of antioxidant components. In addition, the pepper showed significant lipophilic antioxidant activity. Burgers with pepper and plum in different percentages increased the content of phenolic compounds. The ingredients significantly reduced lipid oxidation of the pork burger.

Conclusion:

Natural ingredients increase the content of bioactive compounds when included in pork burgers. The ingredients showed a more antioxidant effect than sulphites in the pork burgers. With these ingredients revalued from by-products, we would obtain natural and healthy burgers.

Valorisation of Hemp Seed Flour for Sustainable High-Moisture Meat Analogues

Özge Güven¹, Ulunay Altanlar¹, **Prof. Dr. İlkey Şensoy¹**

¹MIDDLE EAST TECHNICAL UNIVERSITY

Aim: Meat alternatives, also known as meat analogues, replacers, and substitutes, are gaining attention as they promise to satisfy the dietary needs expected from sustainable alternative protein sources. Hemp seed is an attractive alternative protein source since it is a complete protein source with a complementary balanced amino acid profile. It is a sustainable crop supporting soil health and biodiversity as well as efficient at carbon absorption. These benefits of hemp seed make it an excellent candidate for producing meat alternatives. Among meat alternatives, the extruded plant-based meat analogues capture interest due to their potential to mimic the authentic sensory profile of the meat. In light of this knowledge, this study explores the production of high-moisture meat analogues (HMMA) using hemp seed flour, a by-product of oil extraction, blended with gluten flour through extrusion.

Method: The gluten-hemp seed blends were prepared in predefined ratios (G: H 100:0, 50:50). Meat analogues were produced at high moisture (60%) using a twin-screw extruder with a long cooling die. Two temperature profiles, with end zones at 100°C and 120°C, were used in the barrel. Texture profile and colour analyses and Fourier Transform Infrared Spectroscopy (FTIR) analysis were performed on the extrudates to investigate the samples' physical properties and molecular interactions.

Results: The hardness values increased from around 55 N to 100 N due to added hemp flour, while extrusion temperature did not affect hardness significantly. Colour analysis showed that hemp addition decreased the lightness (L*) values of the analogues from 69-65 to 43-41, showing the darkened colour. FTIR data revealed protein-related molecular interaction changes resulting from extrusion and hemp addition.

Conclusion: The findings of this study support the potential of hemp seed flour utilization in HMMA formulations, offering insights for developing sustainable, consumer-appealing plant-based meat alternatives. Advancing this research will provide knowledge on achieving high-quality, market-ready plant-based meat substitutes that align with contemporary food sustainability goals.

Exploring hydrodynamic-cavitation for citrus waste-valorisation in Malta: from beverage enhancement to potato sprouting-suppression and water-remediation

Dr Georgios Psakis^{1,2}, Dr Frederick Lia^{1,2}, **Prof. Vasilis Valdramidis**³, Prof. Ruben Gatt¹

¹University of Malta, ²MCAST, ³National and Kapodistrian University of Athens (NKUA)

Aim: The endorsement of circular economy, zero-waste, and sustainable-development strategies by the European Union and the United Nations, has encouraged the adoption of non-thermal technologies in both the agro-food and health industries. Whilst integration of such technologies in the processing pipelines of northern European countries has been progressing in fast paces, pathways for their implementation in the food-supply chains of the Mediterranean region appear uncertain or undefined. Considering the fresh orange juice supply chain of the Maltese Islands, we have assessed the usefulness of hydrodynamic cavitation (HC) in the valorisation of orange peel waste.

Method: In detail, we have bench-scale assessed: a) the effectiveness of HC in bioactive compound (total phenolics and flavonoids) extraction from the collected peel of squeezed Navel and Valencia oranges (*Citrus sinensis*) in water (35°C) and 70% (v/v) ethanol (-10°C) over time (15-120 min) relative to conventional maceration at 35°C, b) the antioxidant power of the extracts using the DPPH, ABTS^{••}, and H₂O₂ scavenging assays, c) the capacity of the processed peel to act as a suppressor of potato sprouting and d) the biosorbent potential of the processed peel for copper, nitrate and nitrite binding.

Results: Our work revealed that: a) hot-air drying can be a cost-effective alternative for orange peel waste drying, instead of the more costly freeze drying, b) prolonged HC-assisted extractions in water, at high cavitation numbers, can mechanically damage and/or oxidise the bioactive compounds, with flavonoids and ascorbic acid appearing more sensitive to the treatments, c) cold extractions in 70% (v/v) ethanol, preserve the nature of flavonoids and those organic acids that contribute to increased radical scavenging, and d) HC-processing provides an adequate level of physical peel modification, facilitating its use as a potato suppressant and biosorbent for copper, nitrate and nitrite.

Conclusion: The conducted work provides evidence of the suitability of hydrodynamic cavitation for bioactive compound extraction from orange peel waste in Malta, particularly when coupled with the use of left-over processed peel in potato-sprouting prevention and the construction of biosorbent or biochar fixed-bed columns for the removal of pollutants from irrigation water. This proposed coupling of processes offers a straightforward approach to initiating circular economic practices in Malta, promoting sustainable agriculture.

Utilizing Fruit Pomace and Microalgae for Nutrient-Rich Smoothies: Antioxidant, Protein, and Cholesterol regulation

Miss Ândria Viegas¹, Miss Mafalda Raio¹, Mrs Maria João Alegria², Mrs Anabela Raymundo¹

¹LEAF - Linking Landscape, Environment, Agriculture and Food Research Center. Instituto Superior de Agronomia. Universidade de Lisboa, ²Sumol+Compal Sa

Aim:

Smoothies are blended beverages made from fruits, vegetables and liquids. They're known for their convenience, versatility and health benefits, serving as quick breakfasts, snacks, or post-workout refreshments. Depending on the ingredients, smoothies can offer essential nutrients like vitamins, minerals, fiber, and antioxidants. They can be customized to suit various tastes and dietary needs. This study investigates the formulation and health benefits of three distinct types of smoothies using fruit pomace and microalgae. Each variant is designed to address specific health concerns, including antioxidant support, protein supplementation and cholesterol regulation, while leveraging the nutritional richness of fruit pomace and the unique properties of microalgae.

The initial option centers on smoothies abundant in antioxidants, integrating pear pomace, often discarded during fruit processing, to provide a potent dose of polyphenols, flavonoids and other antioxidant compounds.

The second variant emphasizes protein supplementation, using apple pomace and microalgae. The tests were carried out with two different algae, namely as spirulina and *Chlorella vulgaris* as a source of proteins and minerals.

The third variant aims to contribute to cholesterol regulation; pear pomace known for being rich in fiber was used as well as oats and chia seeds rich in omega 3. These smoothies offer potential benefits for cardiovascular health and lipid metabolism.

Method:

The proximal composition of smoothies (moisture, ash, fat, and carbohydrates) was determined according to standard AOAC methods. Crude protein was quantified by the Dumas method. Total phenolic content was estimated through an adaptation of the Folin-Ciocalteu method and antioxidant activity was measured by two different assays, namely ferric reducing antioxidant power and free radical scavenging activity.

Results:

Pear pomace has been shown to be a source of antioxidant compounds. In this study, pear pomace presented a value of phenolic compounds 1.015 ± 0.04 mgGAE/g fresh weight. The Antioxidant activity was analyzed by the FRAP and DPPH method obtaining a value of 2.327 ± 0.01 and 0.916 ± 0.01 $\mu\text{mol Trolox/g}$ Fresh weight, respectively.

Conclusion:

Effective monitoring of product quality requires understanding their physicochemical attributes, covering rheology, particle size and nutritional composition. This research highlights the health advantages of integrating fruit pomace and microalgae into smoothie and at the same time promoting the circular economy by reducing waste.

Sensory aspects of olive pomace valorisation – identifying key phenolic compounds responsible for bitterness

Yue Ling Wong¹, Dr. Samy Boulos¹, Annette Bongartz², Prof. Dr. Laura Nyström¹

¹Laboratory of Food Biochemistry, Institute of Food Nutrition and Health, Department of Health Sciences and Technology, ETH Zurich, ²Research Group for Food Perception, ZHAW Zurich University of Applied Sciences

Aim:

In olive oil production, 80% of the olive fruit ends up as the by-product olive pomace, a viscous residue that is harmful to the environment if left untreated due to its high content of organic substances. Olive pomace contains a large amount of biophenols, with 98% of the phenolic compounds present in the olive fruit left in the pomace after oil extraction. Given the increasing demand for natural plant-derived food ingredients instead of synthetic ones, olive pomace is a promising source of biophenols that can be valorised for added-value use in food products. However, upon application of olive pomace extracts to food products, a significant impact on flavour due to their characteristic bitter taste can be observed. Hence, investigation of the key bitter phenolic compounds is crucial for development of efficient strategies to valorise the antioxidant potential of olive pomace without compromising on the sensory aspects of the final food product.

Method:

Spanish olive pomace extracts were separated and fractionated under food grade conditions with flash chromatography, using a C18 column and a gradient of H₂O:EtOH. The phenolic compounds across the different fractions were identified with UPLC-DAD-MS/MS, and their bitterness intensity investigated by sensory evaluation with a tasting panel.

Results:

18 phenolic compounds and derivatives were tentatively identified based on comparison with commercial standards or by MS/MS analysis in accordance with literature findings, among which hydroxytyrosol and 3,4-dihydroxyphenyl glycol were the two most abundant phenolic compounds. Upon sensory evaluation, both compounds were shown not to significantly contribute to the bitterness of the olive pomace extract, whereas other minor phenolic compounds including verbascoside and oleacein were identified as key contributors to bitterness.

Conclusion:

Identification of the main phenolic compounds present in olive pomace, as well as the key bitter contributors among them, allows for the optimisation of valorisation of olive pomace. The results of this study have implications for the application of olive pomace biophenols as functional ingredients to food products, where the potential sensory impact of the biophenols must be taken into account.

The grape pomace wastes as a novel for gummy functional food

Dr. Cinzia Mannozi¹, Francesca Pompei¹, Riccardo Marconi¹, Giovanni Caprioli¹, Gianni Sagratini¹, Sauro Vittori¹

¹Chemistry Interdisciplinary Project (CHIP) - School of Pharmacy, University of Camerino

Aim:

Nowadays, the problem of food waste is recognized as a key challenge on the way to sustainable resource management. Particularly much interest is devoted to fruit and vegetable processing due to the abundance of valuable bioactive substances of a health-promoting nature. In this regard, in order to valorize the local producers contributing to fully address the sustainable issues, a functional food (gummy) with some beneficial properties has been developed by using grape pomace waste.

Method:

Functional component of gummy has been obtained by green extraction method such as microwave assisted extraction (540W for 3min), starting from grape pomace of Lacrima di Morro D'Alba variety. Different formulations have been investigated by using different gelling agents (animal gelatin, carragenins and pectin) as well as varying the grape pomace extracts, in order to increase the antioxidant activity of the product formulation. All samples have been analyzed in terms of physico-chemical properties such as colour, structural features, pH, dry matter and A_w and their functionality.

Results:

As regarding the total polyphenols content the formulated gummy showed a slight decrease compared to the grape pomace extract. The product obtained by animal gelatin presented higher firmness compared to plant gelling agents.

Conclusion:

Thus, from a processing waste it is possible to create an innovative healthy and appealing product for the consumer, by reducing food waste promoting a healthier and more conscious lifestyle watching out the environmental issues. This work has been funded by the European Union – NextGenerationEU under the Italian Ministry of University and Research (MUR) National Innovation Ecosystem grant ECS00000041 - VITALITY - CUP J13C22000430001.

Pilot plant extraction of oligo/polysaccharides from cranberry pomace and assessment of their techno functional properties

Master Student Rasha Aleed¹, Research associate Najla Ben Akacha¹, Research Associate Amanda Waglay¹, Research assistant Farnaz Jozedaemi¹, **Professor Salwa Karboune¹**
¹Mcgill University

Aim:

Consumer preference for healthier dietary choices has incentivized food industry to search for novel sources of natural ingredients. Recently, there has been a focus on exploring by-products generated during cranberry juice processing as potentially inexpensive sources of novel functional compounds, including dietary fibers and oligosaccharides. This work aims to investigate the pilot-scale production of oligo/polysaccharides from cranberry pomace by using sequential hot water/alkaline or enzymatic extraction. The structural feature of generated carbohydrates and their techno-functional properties were characterised.

Method:

Cranberry pomace (1.5% w/v, 200 L) was subjected to hot water extraction for 3h at 50°C with constant stirring. The recovered precipitate was then used to extract the oligo/polysaccharides by alkaline treatment (1.23 M, NaOH) or enzymatic treatment (Viscozyme L). All retentates collected from water, alkaline and enzymatic extractions were characterized in terms of recovery yield, carbohydrate content, monosaccharide profile and molecular weight distribution. Several techno functional properties were also assessed including emulsifying, water/oil holding and foaming properties.

Results:

Pilot plant extraction was successfully achieved to produce oligo/polysaccharides rich extracts from cranberry pomace. The recovered carbohydrates upon water, alkaline, and enzymatic extraction were characterized. The carbohydrate yields of 22.3, 18.9, 23.2 and 33.02 % (w/w) were obtained upon water, alkaline and enzymatic (> 5 kDa ; 5 > kDa), respectively. The enzymatic extracts exhibited a predominant presence of hemicellulosic polysaccharides associated with high contents of glucose and mannose, xylose. Conversely, water- and alkaline extracts were predominantly composed of pectic polysaccharides as shown by their high contents in rhamnose, arabinose, galactose and galacturonic acid. The high molecular weight polysaccharides (100-300kDa) were recovered mainly in the alkaline extract, while oligosaccharides (0.5-3KDa) were found in water (53.2%) and enzymatic extract (83.25%). Carbohydrates isolated under alkaline conditions exhibited higher emulsifying properties at concentration 0.01% (911.4 m² /g) and high oil holding capacity (8.2 g/g). Enzymatic-based carbohydrate extracts demonstrated the highest foaming capacity.

Conclusion:

A pilot scale process for isolation oligo/polysaccharides from cranberry pomace was successfully developed using different approaches. The produced carbohydrate extracts exhibited various techno functional properties demonstrating their potential to be used as novel functional ingredients in many food applications.

Green Process for Valorizing Agro-food Residues with High Lignocellulosic Content

Dr. Andrea Bassani¹, Dr. Martina Nardi², Dr. Letizia Crema¹, Dr. Athanassia Athanassiou², Prof. Giorgia Spigno¹

¹Università Cattolica Del Sacro Cuore, Department for Sustainable Food Process (DiSTAS), Via Emilia Parmense 84, 29122 , ²Istituto Italiano di Tecnologia (IIT), Smart Materials, Department of Nanophysics, via Morego 30, 16163

Aim:

This research has been developed in the framework of CYCLEVIT project, which aims to enhance the value of agro-food residues, with high lignocellulosic content, to produce high-value-added products: vitrimers. This work presents a more sustainable process for recovering lignin and cellulose (i.e. the base for vitrimers production) compared to the traditional one which involves acid and alkaline hydrolysis. Such process combines autohydrolysis, to remove the hemicellulose and recover some bioactive compounds, and organosolv to separate lignin from cellulose.

Method:

Different agro-food residues (wheat straw, hazelnut shells, olive seeds cakes, etc.) were analyzed to determine the most suitable in terms of lignocellulosic content. The autohydrolysis optimal condition for lignin and cellulose recovery was identified, based on both previous scientific work and a mathematical model. The solid obtained from autohydrolysis was subjected to the organosolv process which optimal conditions were evaluated by testing different temperatures (180°C-200°C) and solid-liquid ratio (1:10 -1:20 g/ml). The 60 % v/v of ethanol, used for organosolv, was obtained by mixing ethanol with the liquor resulting from the autohydrolysis process.

Results:

The selected residue (i.e. wheat straw) was treated with a combination of autohydrolysis and organosolv. Autohydrolysis was conducted at 190°C for 30 minutes with a solid-liquid ratio of 1:20, based on the results of the mathematical model. After such treatment, cellulose is the main component in the solid (54.33%), with a recovery of 82.50%, the second one is lignin (36.49%) while hemicellulose has been almost completely removed (1.96%), in agreement with the aim of the treatment. The best conditions of organosolv (solid-liquid ratio of 1:20 and 200°C) allows to obtain a lignin recovery of 20.52% with just a slight cellulose degradation (recovery of 91.38%). The acid liquor recycled from autohydrolysis allows a reduction in water consumption of about 36% while also eliminating the need to dispose of acid liquor.

Conclusion:

The new green process presented allows to valorize agro-food residues recovering about 75% of cellulose and 21% of lignin with a water savings of about 36%. However, further investigation is needed for instance regarding the optimal condition for treating other residues or for ethanol recovery.

Using nutritional scores for assessing the quality of dietary lipids from low trophic marine species

Phd Candidate/Researcher Maria Alquiza Medina^{1,2}, Rasa Slizyte¹, Ana Carvajal¹, Eva Falch²

¹Sintef Ocean, ²Norwegian University of Science and Tehcnology

Marine oils contain dietary fatty acids that play a major role in the prevention (and treatment) of life style diseases, such as the cardiovascular diseases. In nature, fatty acids occur in the form of mixtures of saturated fatty acids (SFA), monounsaturated fatty acids (MUFA), and polyunsaturated fatty acids (PUFA), so their nutritional and health benefits should not be evaluated separately. In a world were the population is increasing, efforts are made to investigate which food sources can be important as future essential nutrients.

Aim: The aim of this study was to assess the nutritional value of dietary fatty acids from low trophic marine species and their performance compared to reference seafoods. **Method:** Fatty acid composition of mixed catches of mesopelagic species and individual species of the mesopelagic fishes *Benthoosema glaciale* and *Maurolicus muelleri*, Northern krill and the copepod *Calanus* were analysed. Furhermore, fatty acid compositional data from commercial marine oils from anchovy, cod liver, salmon, and Antarctic krill, were obtained from the Codex guidelines. The quality of dietary fatty acids was assessed using the following scores: PUFA/SFA, thrombogenicity index (IT), sum of eicosapentaenoic acid and docosahexaenoic acid (EPA + DHA), fish lipid quality (FLQ) and the ratio of omega-6 to omega-3 PUFA (n-6/n-3). **Results:** The nutritional quality of marine oils varies widely between species, both between the individual low trophic species and in comparison with commercially available marine oils. The nutritional scores of the oils from the low trophic species were relevant when compared to marine oils from reference seafood. **Conclusion:** This study represents a first attempt to assess the combined effects of dietary lipids derived from low trophic marine species, providing insight into their potential as a marine dietary lipid source. Such an approach goes beyond mere analysis of chemical composition and provides complementary insights that can support more informed decisions regarding food choices for improved nutrition. By considering the broader context of dietary lipid sources, this research aims to contribute to a more comprehensive understanding of marine dietary lipids and their implications for health.

Pulsed electric field (PEF) pre-treatment to protein extraction from pork liver. Influence on technofunctional properties

Dr. Marina Contreras Ruiz¹, Imane Imane Abounnour¹, Jose Benedito¹, Jose Vicente Garcia-Perez, Paola Navarro-Vozmediano¹

¹Polytechnic University of Valencia

Aim:

The aim of this research was to analyze the feasibility of using pulsed electric field (PEF) pretreatment to improve protein extraction from pork liver and its impact on technological properties of protein isolates.

Method:

Protein isolates (PI) were extracted by pH-shift solubilization followed by acid precipitation from pork liver and protein isolates obtained were freeze dried. PEF (1 kV/cm, pulse width 25 μ s and frequency 10 Hz) was applied as a pre-treatment in the raw pork liver. A Box-Benken experimental design (n=3) of 3 factors at 3 different levels was performed by modifying the energy applied in the PEF treatment (0-control, 50, 100 kJ/kg), solubilization temperature (20, 30, 40 °C) and solubilization time (10, 15, 20 min). As response variables, isolate's yield, protein content and technological properties (WHC-Water and OHC- Oil Holding Capacity) were analyzed.

Results:

Protein content in the PI was improved when PEF pre-treatment was applied. Thus, the protein content in control samples (without PEF) was significantly ($p < 0.05$) lower (62.0 ± 2.2 g/100 PI) than in PEF treated but the effect of the energy applied was negligible (73.8 ± 1.9 and 75.8 ± 2.2 g/100 g PI for 50 and 100 kJ/kg, respectively). However, the PI yield was significantly higher ($p < 0.05$) for control samples ($46.5 \pm 1.6\%$) than for PEF treated ones ($27.6 \pm 1.4\%$). Regarding PI technological properties, it was observed that WHC of PI from control samples was lower (2.5 ± 0.1 g water retained/100 PI) than for PEF ones (3.05 ± 0.1 g water retained/100 PI). An opposite tendency was found for the OHC, which was higher (3.3 ± 0.3 g oil retained/100 g PI) in pretreated samples. Solubilization time and temperature did not affect significantly ($p > 0.05$) protein content, protein extraction yield, WHC or OHC.

Conclusion:

PEF pretreatment during protein extraction improved the protein content of the protein isolate; nevertheless, reduced the protein extraction yield. Furthermore, OHC was enhanced in the protein isolate obtained with PEF pretreatment but the opposite effect was found for WHC. Future studies should elucidate the mechanisms linked to the PEF application on the pork liver which are responsible of the differences observed in the protein isolates analysed in the present work.

Techno-functional behavior of legume grass protein concentrates within model food systems

Esteban Echeverria-jaramillo¹, Mette Lübeck², Simon G Echers², Anders K Jørgensen², Tuve Mattsson², Peter S Lübeck³, Vibeke Orlien¹

¹Department of Food Science, University Of Copenhagen, ²Department of Chemistry and Bioscience, Aalborg University, ³BiomassProtein APS

Aim: Protein concentrates extracted from upcycled legume grass (LGCs) represent a promising ingredient for the green transition of the food infrastructure. Despite their remarkable functional properties observed in laboratory settings, their behavior under real food conditions remains unknown. Therefore, this study aims to evaluate the techno-functional properties of LGCs across four different model systems varying in pH and ionic strengths, to identify their optimal applications in product formulations.

Method: LGCs were produced through an ultrafiltration process and dried via spray drying (SD). Three variants were generated: G1 (no additive/pilot-scale SD), G2 (antifoaming added during filtration/pilot-scale SD), and G2F (antifoaming added during filtration/industrial-scale SD). The foaming and gelling properties of these LGCs were evaluated across five model systems. These included lab conditions (1% w/w powder in deionized water at native pH and ionic strength) and combinations of pH 4 and 7, along with low (0.02 M) and high (0.2 M) ionic strength, with a protein concentration of 1% (w/w) hydrated in tap water.

Results: The functionality of G1 was inferior compared to G2 and G2F. The latter two showed no significant differences, indicating that the scale of the drying process has no effect as long as temperature conditions are similar. Foaming capacity of the three LGCs ranged from $106 \pm 4\%$ to $122 \pm 2\%$ irrespective of pH or ionic strength. However, foam stability was notably improved in low pH systems with G2 and G2F maintaining ~80% of their original volume after 15 minutes. The high stability at low pH corresponded to the lowest protein solubility of LGCs, giving new insights into LG proteins' foam mechanism. Regarding gelling, an opposing phenomenon occurred: at pH 4, protein precipitated limiting its participation in the gelling process.

Conclusion: The addition of antifoaming during LGC production did not affect functionality. LGCs' functionality was controlled by the pH of the system, while ionic strength did not play a major role. These findings provide insights for determining the optimal application of LGCs in food products to maximize their performance. Future research should design more complex food systems incorporating components such as lipids and heat treatments.

Exploring artichoke by-products for sustainable kombucha innovation: a circular economy approach

Ph.D. Olaia Estrada¹, Telmo Puente¹, Ph.D. Iñaki Diez-Ozaeta¹, Blanca del Noval¹

¹Bcc Innovation, Technology Center In Gastronomy, Basque Culinary Center

Aim: The kombucha market has witnessed a significant increase in demand. Currently, this drink has become one of the most popular low-alcoholic fermented beverages. While black and green tea varieties have traditionally served as substrates for preparing kombucha, a wide range of alternative substrates are nowadays investigated.

Artichoke is traditionally consumed in the Mediterranean diet in different popular preparations. During the artichoke processing, the residues, principally external leaves or stems, represent approximately 60–80% of the total harvested plant material. The aim of this study was to explore artichoke by-products as potential substrate to develop innovative fermented beverages.

Method: Kombuchas were prepared adding commercial SCOBY (Symbiotic Consortium of Bacteria and Yeasts) to a sugared infusion ($70 \text{ g sucrose L}^{-1}$) prepared with dried external artichoke leaves (1.3% w/v). Kombuchas were kept at room temperature ($23 \pm 2 \text{ }^\circ\text{C}$) during 7 days. Afterward, the kombuchas were filtered, bottled, and kept refrigerated at $4 \pm 1 \text{ }^\circ\text{C}$ during 4 months. Total acidity (TA), pH, total soluble solids (TSSs), sucrose, fructose, and glucose were analysed at 0, 1, 7, 30, 60 and 120 fermentation days.

Results: During the initial 7 days of fermentation, significant changes in composition occurred. The pH values decreased from 5.89 ± 0.05 to 3.52 ± 0.03 , while the total acidity increased from 0.027 ± 0.003 to $0.292 \pm 0.008 \text{ g/100 mL}$, respectively. There was a rapid increase in glucose and fructose content (25.4 ± 0.2 and $22.5 \pm 0.3 \text{ g/L}$, respectively), while sucrose was completely consumed. After bottling, there were no significant changes in the pH, TA, and glucose content. However, fructose decreased to $9.6 \pm 0.3 \text{ g/L}$. TSSs values decreased significantly ($p < 0.001$) during all studied period.

Conclusion: This study highlights the feasibility of utilizing artichoke by-products in kombucha production, contributing to the development of new varieties of this popular fermented beverage within the circular economy framework. The stability of pH, total acidity, and glucose content after bottling suggests the potential for shelf-life preservation and aging.

Keywords: artichoke by-products, circularity, new drinks, kombucha, fermented beverage, storage

LCA of waste streams from tomato cultivation and production

Andreas Gess¹, Mecit Oztop¹, Özlem Özmutlu Karslioglu¹, Simon Dirr¹, Lisa Ziegltrum¹

¹Greensurvey GmbH

Aim:

Tomato plays a pivotal role in the Mediterranean diet. The by-product of tomato processing, known as tomato pomace, has garnered attention for its nutritional and textural qualities, making it suitable for both feed and food applications. Despite its potential, there is currently a lack of defined technologies for scaling up the production of these products for widespread use. Due to its protein content, tomato pomace presents an opportunity to serve as a valuable protein source. Additionally, tomato leaves yield a relevant amount of protein to be processed into protein powder. While both raw materials show a high potential for converting a waste material in a valuable substance, studies are hardly found or still non-existent. Within the project ProxIMed, various technologies to extract proteins from tomato pomace and tomato leaves are tested on their technological feasibility but also on their ecological sensibility. For this purpose, a Life Cycle Assessment (LCA) of the process schemes is conducted.

Method:

A classic LCA is conducted based on the ISO norms 14040/14044 for protein products made from tomato pomace and tomato leaves. Since one is a by-product while the other is a waste, allocation of environmental impacts is the central issue in this study.

Results:

The LCA results are focussing on impacts on climate change, resource use, land and water use. A special focus is set on the allocation of the impact, meaning the share of impacts that are accounted for the various products within the tomato production.

Conclusion:

The presented study gives an overview of the ecological aspects of protein products made from tomato pomace and tomato leaves and their potential of contributing to the minimization of the environmental impact caused by food provision. It also presents one of the first studies ever to shine a light on making-use of the waste streams of tomato cultivation.

Evaluation of Ultrasound-Assisted Extraction of lemon seed extracts using ethanol as green solvent

Mayra Chalapud^{1,2}, Agustin Benestante^{1,2}, **Dr. Anabella Giacomozzi**³, Jose Vicente Garcia Perez³, Jose Benedito³, Maria Elena Carrin^{1,2}

¹Departamento de Ingeniería Química, Universidad Nacional del Sur (UNS), ²Grupo de Ingeniería de Alimentos, Planta Piloto de Ingeniería Química (PLAPIQUI) , ³Instituto de Ingeniería de Alimentos-FoodUPV. Universitat Politècnica de València

Aim:

Ultrasound-assisted extraction (UAE) is an alternative and environmentally friendly technology usually resulting in reduced time, energy and solvent consumption, and unit operations compared to conventional extraction techniques. Lemon seeds (LS) constitute a valuable by-product rich in biocompounds. Ethanol is an alternative among green solvents, capable of solubilizing different interesting components, such as oil and carbohydrates, among others. The aim of this work was to analyse the effect of time, temperature, and solvent:seed ratio (SSR) on the total extraction yield (TEY) and hexane-insoluble material yield (HIMY) obtained from UAE of lemon seed using ethanol as solvent.

Method:

LS were dried ($7.88 \pm 0.15\%$ d.b. moisture content) and grounded. Soxhlet extraction (6 h, SSR=10 ml/g) and UAE were evaluated using ethanol 96% as solvent. UAE was performed using a probe operated by pulses (20 kHz, 1 s on/off, 40% amplitude). Ethanolic extracts were fractionated by liquid-liquid separation using hexane, obtaining two fractions: hexane-soluble and hexane-insoluble. The total extraction material was calculated as the sum of these two fractions. A central composite rotatable design was used with three independent factors: time (6, 13, and 20 min), temperature (35, 42.5, and 50 °C), and SSR (5, 10 and, 15 ml/g), having TEY and HIMY (% d.b.) as responses. Optimal UAE conditions were determined using Response Surface Methodology.

Results:

TEY and HIMY obtained by Soxhlet were $39.01 \pm 1.76\%$ and $11.15 \pm 1.42\%$, respectively. The UAE results showed that the three factors had a significant effect ($p < 0.05$) on HIMY, whereas the time was insignificant for TEY ($p > 0.05$). When the response TEY was maximized and HIMY minimized, the optimal conditions were: 6 min, 35 °C, and 15 ml/g, with TEY=13.44% and HIMY=3.00%. On the other hand, when TEY and HIMY were maximized, the results were: 20 min, 50 °C, and 15 ml/g, with TEY=14.51% and HIMY=5.34%.

Conclusion:

Although the TEY and HIMY values obtained with UAE were lower than those achieved with Soxhlet extraction, it is noteworthy that extraction of LS compounds can be reached in shorter times and lower temperature using UAE. Future research into environmentally friendly technologies and green solvents for by-product valorisation is greatly encourage by these studies.

Recovery of bioactive compounds from tomato pomace using vegetable oils as green solvents: Bioaccessibility study

Ioanna Thanou¹, Dr Maria Katsouli¹, Rouainta Alsaoua¹, Dr Athina Ntzimani¹, Dr Maria Giannakourou¹, Prof Petros Taoukis¹

¹National Technical University Of Athens

Aim:

In the context of utilizing food industry by-products, the retrieval of significant substances with high-added value has become a priority. Currently, using organic solvents as extraction media is a common practice for the recovery of bioactive compounds from food industry by-products. However, the increasing demand for eco-friendly, 'green' solvents has brought vegetable oils to the forefront as an ideal alternative, both facilitating effective extraction and enhancing the oils themselves. Tomato pomace (by-product of tomato industry) is rich in antioxidant compounds, such as lycopene, which offer significant health benefits. Therefore, the aim of this study was to optimize the extraction of lycopene using vegetable oils as solvents and to determine the bioaccessibility of lycopene in each oil.

Method:

For the extraction of lycopene from tomato pomace, vegetable oils such as olive, olive kernel, canola, corn, and sunflower oils were used as alternative solvents. The kinetics of lycopene extraction were investigated at a constant temperature of 50°C for different liquid-to-solid ratios (5:1-30:1). The extraction yield and the oxidation of the fatty components were determined. Subsequently, the enriched oils were tested in order to evaluate the effect of the type of oil on the bioaccessibility of lycopene. For the determination of bioaccessibility, the static INFOGEST *in vitro* protocol was applied.

Results:

The results indicated that the maximum lycopene recovery was recorded at 50 °C, using a 5:1 oil to pomace ratio, after 24 hours of extraction. Regarding bioaccessibility, olive oil, canola oil and olive pomace oil exhibited similar behavior, presenting the highest bioaccessibility (22.1%, 21.7% and 21.3%, respectively). This can be attributed to their high content of oleic acid (C18:1), which has a positive effect on bioaccessibility due to its long carbon chain length and monounsaturated nature. Corn oil and sunflower oil demonstrated lower bioaccessibility (15.6% and 14.2% respectively), possibly due to the oxidation of their polyunsaturated fatty acids during digestion.

Conclusion:

Vegetable oils can serve as efficient alternative solvents for extracting valuable bioactive compounds from food industry by-products, leading to enriched foods with high bioaccessibility. This approach aligns with the growing trend towards sustainable and health-conscious practices in the food industry.

An efficient extraction process focusing on the recovery of flavonoids from *Cannabis sativa*

Emmanouil Vachatsakis¹, Georgios Theocharis¹, Dr Dimitrios Tsimogiannis¹, Dr Maria Giannakourou¹

¹National Technical University Of Athens

Aim:

Hemp or industrial cannabis (*Cannabis Sativa L.*) belongs to the Cannabaceae family. Cultivation of the plant for food applications had been restricted in many EU countries for decades, due to its minor content in the psychoactive component THC. Recently, the EU legislation was reformed and numerous hemp products have been developed. Also, the scientific research on the compounds of hemp has drastically increased. Apart from essential oil and cannabinoids, the plant contains significant amounts of flavonoids belonging to the subgroup of flavones. Among them, two prenylated forms of the flavone diosmetin, cannflavin A and B, could be distinguished. The compounds have demonstrated bioactivities such as anti-inflammatory and neuroprotective effects. The scope of the current research deals with the efficient and applicable in large scale recovery of cannabis flavonoids as an additional extractable fraction of bioactive compounds from hemp, beyond essential oil and cannabinoids.

Method:

Organically cultivated hemp was subjected to a series of sequential extractions with acetone and water so as to recover and characterize the main non-polar and the polar fractions of the plant. Additionally, and only for analytical purposes, methanol was used to extract residual compounds belonging to all polarities. The extracts were characterized using chromatographic methods as well as widely used protocols such as Folin-Ciocalteu and DPPH, so as to develop an extraction method applicable in large scales, aiming at recovering the majority of flavonoids from the plant.

Results:

The experimental results indicated that, while the majority of hemp flavonoids can be recovered by water extraction, the cannflavins, cannot be extracted either by acetone or water individually. Only methanol recovered the compounds efficiently. In order to avoid the use of toxic methanol, or ethanol that would increase the complexity of the process forming azeotropic mixtures, the use of acetone-water mixture was examined as an alternative for the extraction of cannflavins that lead to a successful outcome.

Conclusion:

A series of successive acetone, aqueous acetone, and water extractions can lead to the exhaustive recovery of all hemp compounds including the highly bioactive cannflavins for use as ingredients for pharmaceutical, food, supplements and cosmetic preparations.

Sustainable production of carotenoids from red yeast using soybean waste

Phd Candidate Jiaqi Liang¹

¹Nanyang Technological University, ²Agency For Science, Technology And Research

Aim:

Okara, the pulp that is left behind from soymilk and tofu production, is a common waste in Asia. Despite being typically discarded or used in low-value applications, its nutrient-rich composition and high nitrogen content make it an ideal candidate for microbial biovalorisation. Thus, this study aims to explore the upcycling of okara using red yeast to produce high-value carotenoids.

Method:

Okara was fermented with a food-grade fungus to enhance nutrient bioavailability. An aqueous extract of the soluble nutrients from fermented okara was used to replace the nitrogen component of traditional YPD media for yeast culture. *Xanthophyllomyces dendrorhous*, a red-coloured yeast known for its ability to utilise alternative substrates and to produce the high-value carotenoid astaxanthin, was cultured for five days in the okara media. To further improve nutrient availability, co-culture with a nitrogen-fixing bacteria *Paenibacillus polymyxa* was tested. Cell growth, beta-carotene and astaxanthin production were measured throughout the culture duration to determine the performance of the okara media relative to the conventional YPD control. Differences in cell growth and carotenoid production were characterised by metabolomics studies.

Results:

The okara media supported enhanced cell growth and carotenoid production in the red yeast. Compared to the YPD control, astaxanthin levels were found to be two-fold higher by the fifth day of culture. When co-cultured with *P. polymyxa* in okara media, the total carotenoid content increased two-fold, and the astaxanthin content increased six-fold relative to the YPD control. Metabolomics studies revealed a shift in metabolic flux from a competing fatty acid biosynthesis arm to favour carotenoid biosynthesis instead in okara media.

Conclusion:

Okara extract emerges as a cost-effective and sustainable alternative to traditional microbiological culture media. It not only enhances growth but also significantly improves astaxanthin production in the red yeast *X. dendrorhous*. This presents a promising solution for managing the abundant soybean waste in Asia and contributes to the advancement of a circular economy.

Green extraction of inulin from acai (*Euterpe oleracea*) industry residue

Gabriel Sthefano Lourenço Pereira¹, Patricia Tonon de Souza¹, Renata da Silva Magalhães¹, Antonio Jose de Almeida Meirelles¹, Guilherme Jose Maximo¹, Eduardo Augusto Caldas Batista¹, Klicia Araujo Sampaio¹

¹Universidade Estadual De Campinas

Aim:

In recent years, the growing demand for sustainable practices in the food industry has driven the search for more efficient ways to manage the waste generated during the production process. Byproducts of tropical fruit processing, such as açai (*Euterpe oleracea* Mart.), emerge as a rich source of bioactive compounds and functional ingredients. The significant quantity of these residues, particularly in the form of seeds after pulp processing, presents an opportunity for valorization and the development of value-added ingredients, such as inulin. This study aims to investigate the optimal conditions for efficiently extracting inulin from açai seeds.

Method:

The açai seeds were acquired by Raízes do Açai company, subsequently dried at 60°C and ground. Then, nine extraction runs were conducted using water as the solvent, varying the temperature from 50 to 90°C and the solid-solvent ratio from 1:10 to 1:2 (w/v) for thirty minutes under magnetic stirring. The resulting extracts were filtered and subjected to quantification of inulin content using an ion chromatograph. The best extraction condition was qualitatively evaluated by FTIR, comparing it to the commercial chicory inulin standard.

Results:

The nine extraction runs yielded varying amounts of extracted inulin from the açai seeds. Analysis revealed that extraction run number 6 (90°C and 1:5 solid-solvent ratio) resulted in the most efficient extraction of inulin (19.03 ± 0.30 g/100g). Runs with higher temperatures tended to exhibit improved extraction efficiency, and, despite the expectation of higher inulin content with increasing solid-solvent ratio, runs with medium proportions exhibited better inulin yield. This observation suggests that the difficulty in achieving adequate stirring may have limited the extraction efficiency. Furthermore, qualitative evaluation by FTIR revealed that the inulin extracted from run 6 closely matched the spectral characteristics of the commercial chicory inulin standard.

Conclusion:

This study successfully investigated the optimal conditions for the efficient extraction of inulin from açai seeds. Run 6 emerged as the most promising condition, yielding the highest content of extracted inulin. Additionally, qualitative analysis by FTIR demonstrated that the extracted inulin closely resembled the commercial chicory inulin standard, indicating its quality and potential for application in various food formulations.

Analysis of the composition of hydrolates and their application as preservatives for fresh meat

PhD María Jesús Martín Mateos¹, Phd Jonathan Delgado Adámez¹, Susana García Torres¹, Phd Francisco Vázquez Pardo¹, María Amparo Cabeza de Vaca Molina¹, Phd M.Montaña López Parra¹, PhD Rosario Ramírez Bernabé¹

¹Cicytex

Aim:

Hydrolates are the liquid residue obtained from the process of extracting essential oils from aromatic plants by steam distillation. Essential oils possess antimicrobial and antioxidant properties, but they have very intense aromas. However, traces of essential oil may be present in hydrolats, and their aroma is less intense. The main use of hydrolats is cosmetic, but, due to their properties, they could have different applications in the food industry. In addition, consumers and the meat sector are demanding the production of more natural, healthier, additive-free, and more sustainable meat products. For this reason, the aim of this study was to evaluate the bioactive composition and the antioxidant and antimicrobial properties of hydrolats from three aromatic plants, and to investigate their ability to preserve fresh meat products.

Method:

Hydrolates were obtained at CICYTEX from three aromatic plants (Origanum, Thymus and Lavandula), and their total phenolic compounds content (PCC), the antioxidant and antimicrobial activity were evaluated. Their potential to preserve fresh meat products was evaluated. For this purpose, pork loin steaks were marinated for 24 hours in the hydrolate solutions. Microbiological counts, instrumental color and lipid oxidation were analysed in the pork loin.

Results:

The PPC was significantly higher in the oregano hydrolat, while it was very low in the lavender hydrolat. Similar results were obtained for hydrophilic antioxidant and antimicrobial activity, where the highest values were obtained in oregano hydrolat. The marinate process in hydrolates increased the lightness (CIE L*) values and reduced the redness (CIE a*) values compared to the control (non-marinated pork). Hydrolates were not effective to prevent microbial development in the marinated pork samples, only oregano hydrolat presented a slight antimicrobial effect. No effect was observed on the lipid oxidation either.

Conclusion:

Oregano hydrolat had higher content of phenolic compounds as well as higher antioxidant and antimicrobial activity than the other hydrolates studied. Therefore, this hydrolate was the most effective to prevent lipid oxidation and microbial development in pork loin steaks, so it could be utilized as a marinate solution and it could be a promising strategy to increase the shelf-life of meat.

Boosting biorefinery protein production from green leafy biomass

Mr. Waleed Mlook Waleed Mlook¹

¹Swedish university of agricultural science

Aim:

The global demand for protein is projected to increase by 2050, driving interest in alternative sources such as plant-based proteins. Green leafy biomass, rich in Ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBisCO), presents a potential protein source. However, limited protein extraction rates challenge the economic sustainability of its utilization. This study aims to enhance protein extraction from green biomass, particularly pulp fractions, by improving separation efficiency, exploring extraction factors. Ultimately, it seeks to support sustainable food and feed production to meet growing protein needs.

Method:

The study explored processing of lucerne, at pilot and lab scales. Pilot-scale operations involved washing and pressing with subsequent freezing, while lab-scale investigations focused on the impact of freezing on protein extractability, applying diverse pre-treatment methods like water, ice, NaOH, SDS solutions, enzyme-assisted extraction and sequential pressing. Analytical procedures assessed dry matter and nitrogen levels, with rigorous data analysis to determine mass balances, nitrogen content, and nitrogen flows from pulp (P) to repressed pulp (PR), green juice (GJR) and green pellet (GP).

Results:

Water showed %N flow ranging from 53.9±2.9 to 70.4±3.8 and 34.8±0.8 to 35.4±1.6, respectively, from PtoGJR and PtoPR. NaOH solutions, especially at 1% concentration, yielded the highest %N flow (89.3±5.2) from PtoGJR. SDS solution at 0.5% concentration resulted in a notable %N flow of 95.9±1.9 from PtoGJR SDS. The nitrogen transfer is influenced by material status (fresh/frozen) and pulp: water ratios, with higher ratios generally leading to increased %N flow. Frozen material tends to have higher %N flow compared to fresh material across all ratios. Enzyme concentration variations did not significantly affect %N flow. However, using repressed pulp and a 1:6 water ratio during enzymatic treatments showed %N flow 51.8±1.0 from PtoGJR and PtoPR and resulted in the highest %N flow from GJR to GP (75.1±5.2).

Conclusion:

In conclusion, the NaOH 1% and the SDS 0.5% concentration emerge as promising methods for enhancing protein extraction from lucerne biomass. Further optimization of these techniques, along with exploration of enzymatic treatments using repressed pulp and specific water ratios, holds potential for advancing sustainable protein production from green biomass sources.

Innovative Valorization of Oat By-Products: Enzymatic Extraction and Purification of Proteins for Sustainable Food Applications

Daniel Probst¹

¹Th Owl

Aim:

Food loss and waste are significant challenges to the sustainability of food systems. This study aims to explore innovative approaches for reducing food waste and valorizing side-streams, focusing specifically on oat okara, a by-product of oat milk production. The aim is to extract and purify proteins from oat okara through enzymatic processes, with the ultimate goal of creating a sustainable protein source for various applications.

Method:

Oat okara was subjected to enzymatic treatment and hydrolysis using a pressure filter press. Enzymatic processes were optimized to achieve high protein yields with purity exceeding 80%. The hydrolysate was then concentrated using a rotary evaporator and purified through membrane filtration to remove impurities and smaller molecules.

Additionally, a chromatography system was employed to further purify the protein fractions and isolate individual peptides. This comprehensive approach ensures the extraction of high-quality proteins suitable for various applications.

Results:

The enzymatic extraction and purification process yielded proteins with high purity (>80%) from oat okara. The pressure filter press efficiently facilitated enzymatic hydrolysis, resulting in a hydrolysate rich in protein content. Subsequent concentration and purification steps effectively removed impurities, enhancing the quality of the protein extract.

Furthermore, chromatography enabled the isolation of individual peptides, further enhancing the value-added potential of the extracted proteins.

Conclusion:

This study demonstrates the feasibility of enzymatic extraction and purification of proteins from oat okara, a by-product of oat milk production. The extracted proteins hold promise as sustainable alternatives for various applications, including food and cosmetic industries. By valorizing side-streams and reducing food waste, this approach contributes to the development of more sustainable food systems.

Upcycling of tomato peel as a raw material for the extraction of bioactive compounds

Josipa Dukić¹, Marinela Nutrizio¹, Filip Klemen¹, Mecit Halil Oztop², **Professor Anet Rezek Jambrak¹**

¹University of Zagreb, Faculty of Food Technology and Biotechnology, ²Middle East Technical University

Aim:

Tomato peel is a by-product of the food industry, which is produced during the manufacture of tomato products. It is a good source of bioactive compounds that have beneficial effects on human health. With population growth and the increasing demand for food, the utilization of such a high-quality by-product is extremely important.

Method:

In order to utilize the by-products, the aim of this work was to investigate the influence of the nonthermal extraction method - high-power ultrasound (US) on the physicochemical parameters of dry tomato peel extracts. Compared to the nonthermal method, a thermal extraction method was applied at a temperature of 60°C, and deionized water, 25, and 50% ethanol solutions were used as extraction solvents. From the physical parameters, changes in pH and electrical conductivity (EC) values were observed. In terms of chemical parameters, changes in flavonoid content (FC) and antioxidant activity (AA) were observed (DPPH/ FRAP method).

Results:

Significant differences in pH values between the two extraction methods were not observed. However, a statistically positive influence of ethanol content on the changes in pH values was found for both methods ($p < 0.05$). A similar trend was observed in the EC results, with a difference in the thermally-treated samples. Regarding chemical parameters, a higher yield of FC was found in the US-treated samples. In particular, on the first day of storage, at an amplitude of 75%, a treatment time of 9 minutes and a 50% ethanol solution as extraction solvent, the highest yield of FC was 3.21 ± 0.01 mg rutin/g_{d.m.} (2.07-3.82 times higher yield compared to the yields obtained by the thermal extraction method). Regardless of the used extraction method, a decrease in FC was observed in the samples after 7 days of storage (+4°C). In addition, higher AA values were observed with the FRAP method regardless of the extraction method.

Conclusion:

The obtained results are promising and can serve as a guide for other research that involve the use of nonthermal extraction techniques for the extraction and isolation of bioactive components from by-products and/or waste from the food industry.

Biorefinery Approach for the Recovery of Ingredients from Waste Brine Obtained from Ham Production

Mr. Anim Ujong^{1,2}, Dr. Joncer Naibaho¹, Prof. (Dr) Brijesh Tiwari¹, Mr. Shay Hannon¹, Dr. Uma Tiwari²
¹Teagasc Food Research Centre, Ashtown, ²School of Food Science and Environmental Health, Technological University Dublin

Aim:

Meat processing operations generate large quantities of wastewater streams, which pose a costly challenge for treatment. However, these waste streams, despite undergoing wastewater treatment, still contain considerable amounts of valuable ingredients and provide an important opportunity for waste utilization. The aim of this study was to identify suitable solvent options for efficient ingredient recovery from waste brine generated from the ham production system.

Method:

The waste brine samples was collected from a local ham processing facility in Ireland and analyzed for BOD and COD. The brine was thereafter freeze-dried for compositional analysis e.g. fat, ash, and protein content. The brine was treated with five different types of solvents (ethanol, NaOH, acetic acid, lactic acid, and citric acid) and heated at 60°C for 15 min, centrifuged to obtain solid residue and freeze dried. FTIR spectroscopy was used to assess the secondary structure of proteins in the freeze dried precipitates.

Results:

Brine waste analysis revealed high levels of BOD (6254 mg/L O₂) and COD (21680 mg/L O₂) exceeding the permissible limits set by the Environmental Protection Agency prior to discharge. The freeze-dried brine contained ~12% protein, ~0.96% fat, and ~8% ash. Ethanol produced the highest precipitate yield (3.41%), followed by NaOH (3.31%) while acid precipitates yielded similar results (2.71-2.92%). The precipitates contained high protein contents (35.07-47.11%), with citric acid yielding the highest protein content (47.11%) and ethanol the lowest (35.07%). No significant difference ($p > 0.05$) was observed in the protein content and recovery yield precipitates from acids. FTIR analysis indicated minor alterations in the secondary structure of the protein, with β -sheet and α -helical structures being dominant, indicating that the precipitates may have structural integrity. Furthermore, the results revealed various functional groups of organic compounds such as N-H or O-H stretching, C=O stretching, C-O stretching, and potentially phosphodiester linkages. Proteins recovered could potentially be utilized for various agri-food applications.

Conclusion

Citric acid seems to be the most suitable solvent for recovering ingredients from waste brine. The considerable protein and fat content in the waste brine and precipitates suggests the opportunity for resource recovery while reducing the usage of wastewater treatment systems.

Comparison of the physicochemical and rheological properties of fibres extracted from agri-food by-products against methylcellulose.

Whitney Vale-Hagan¹, Dr Eoin Cunningham¹, Professor Dimitris Charalampopoulos², Dr Tassos Koidis¹

¹Queen's University Belfast, ²University of Reading

Aim: Our exploration of agricultural by-products reveals a range of fibres that possess their own unique characteristics and physicochemical compositions. These fibres have varying physicochemical and functional properties that requires further investigation. Therefore, the study was aimed at extracting and assessing the binding efficiency of dietary fibres extracted from nine agri-food by product sources as an alternative to an industry standard food binder which is methyl cellulose. We aim to expand our understanding of the functional potential of these by-products in food applications specifically plant-based foods.

Method: In this study the physicochemical and rheological properties of dietary fibres extracted from nine different sources namely apple pomace, wheat straw, hemp fibres, oat hulls, oat bran, pumpkin seeds, mushrooms compost, and coffee silverskin were compared to methyl cellulose. The water holding, water binding, swelling and oil holding capacities, rheological and thermal properties, structural characteristics (SEM, FTIR and NMR) were characterised.

Results: Oat hulls (65.82%) and hemp fibre (69.63%) contained the highest DF contents whilst apple pomace (19.52%) and oat bran (28.03%) had the lowest DF content. OPDF exhibited the highest water binding (0.94 g/g) and swelling capacities (13.85 mL/g) whilst MC and AC-APDF observed the highest water holding capacity (12.74 g/g) and AL-APDF with the highest oil holding capacity (8.53 g/g). Only MC exhibited an excellent emulsifying property with no serum layer observed. However, AL-APDF and OPDF showed great emulsification potential. All DF samples and MC exhibited shear thinning behaviours with increasing shear rates. There was a correlation between the apparent viscosity of the DF solutions and the viscoelastic properties. The thermal properties shows that the extracted DF samples were all amorphous in nature with varying microstructures observed under the scanning electron microscope. The FTIR spectral analysis showed that all DF samples contained characteristic peaks of polysaccharides.

Conclusion: The findings indicate the potential fibres have as a functional ingredient in the food industry. Through further investigation and modification, we can enhance and uncover the depths of these fibres' potential as binding agents in plant-based food processing.

Impact of particle size reduction of plant-based sidestreams on pectin extraction yield and pectin structure

Elien De Laet¹, Tom Bernaerts¹, Prof Ann Van Loey¹

¹KU Leuven

Aim:

The food industry produces large amounts of plant-based side streams, for which valorization is needed in order to move towards a more circular bio-economy. Plant-based sidestreams from dicotyl plants contain pectin, a cell wall polymer that is often used as a functional ingredient in the food industry. Current study aimed to investigate the impact of particle size reduction of the biomass and of the fraction mechanism on pectin extraction yield and pectin structural characteristics.

Method:

Three biomasses with an increasingly complex composition, being carrot pomace, broccoli stems and pumpkin pomace, were included in this study. For all biomasses, high shear mixing, high pressure homogenization (HPH) and ball milling (BM) were applied as particle size reduction techniques. Microstructural characterization was done in terms of particle size distribution and cryo-SEM. Apart from determination of the pectin extraction yield and pectin purity after an acid extraction process, the pectin structure was characterized in terms of monosaccharide composition, degree of methylesterification and molecular weight distribution.

Results:

All three particle size reduction techniques resulted in a significant decrease of the median particle diameter. As comparable particle size distributions but different microstructures were obtained after HPH and BM, the effect of the fracture mechanism could be evaluated while excluding the effect of particle size. After acid pectin extraction a comparable and significantly higher extraction yield, compared to the non treated sample, was obtained, indicating that particle size reduction resulted in an increased extraction efficiency. Moreover, it was observed that the fracture mechanism had no substantial influence. Overall, the different particle size reduction techniques had no clear impact on the structural characteristics of the extracted material, and the main difference between the different biomasses was the extent of protein and starch co-extraction during the pectin extraction process.

Conclusion:

The results in this study indicate that both HPH and BM can be used as particle size reduction techniques to increase the pectin extraction efficiency and that the efficiency increase is biomass dependent. The comparable molecular structures suggest that these samples will most probably also have comparable functional properties, which should be confirmed in follow-up experiments.

Determination of Bioactive Properties of Sprouted Dry Faba Bean Protein Extract

Determination Of Bioactive Properties Of Sprouted Dry Faba Bean Protein Extract Fatma

Bozanoglu¹, Dilara Ozcan Ozcan², Gokhan Durmaz¹, Mecit Halil Oztop²

¹Inonu University, ²Middle East Technical University

Aim:

Proteins are essential in food products, and the demand for plant-based proteins is increasing due to their high availability, low cost, and nutritional value. Faba bean (*Vicia faba* L.) has attracted attention among plant protein sources due to its high protein content and environmental sustainability advantages. However, the presence of antinutrients limits the nutritional value of legumes. Sprouting is a food processing technique used to improve the nutritional quality of legumes by reducing the content of antinutritional factors. This study aims to determine the efficiency and bioactive properties of sprouted protein extracts and the effects of the sprouting process on protein quality and potential food applications, which are considered crucial for the development of protein-based food products.

Method:

Dry beans were sprouted at room temperature (25±3 °C) in a moist environment free from daylight and protein extraction was performed by alkaline-isoelectric precipitation method. Protein content was determined by Kjeldahl method and soluble protein by Lowry procedure. Phenolic content, antioxidant activity and color characteristics were also determined.

Results:

The soluble protein content was higher in sprouted faba bean protein extract (SFP) (29.36%) than in dry faba bean protein extract (DFP) (23.27%). The total phenolic content of DFP was 90.78 mg GAE/100 g, while this amount was 157.17 mg GAE/100 g in SFP. There was a remarkable increase in the total phenolic content with sprouting process. The highest values of DPPH (224.79 mg Trolox/100 g) and ABTS (647.66 mg Trolox/100 g) were determined in the protein extract obtained from sprouted faba bean. Color analysis indicated that SFP was relatively darker than DFP, which may be attributed to the increased phenolic compounds during the sprouting process.

Conclusion:

The sprouting process increases the digestibility of proteins by activating enzymes that hydrolyze proteins and changes the protein content and amino acid composition. In addition, phenolic compounds and antioxidant activity values of sprouted samples were also observed to be high. Sprouting increases the nutritional value and bioavailability of protein, which is important for developing sprouting techniques and optimizing protein-based food products. This method can improve the bioactive and functional properties of plant proteins.

Bioactive extracts from ozonated residues of interest in food-related applications

Ms. Mahrokh Ebrahimi^{1,2}, Dr. Antonio Martínez-Abad^{1,3}, Dr. Ana-Rosa Ballester⁴, Dr. Victor Acha², Dr. Thierry Aussenac², Dr. Amparo López-Rubio^{1,3}

¹Food Safety And Preservation Department, Institute Of Agrochemistry And Food Technology (iata), Csic, ²Institut Polytechnique UniLaSalle, Université d'Artois, ULR 7519, 19 rue Pierre Waguet, BP 30313, ³Interdisciplinary Platform for Sustainable Plastics towards a Circular Economy—Spanish National Research Council (SusPlast), CSIC, ⁴Food Biotechnology Department, Instituto de Agroquímica y Tecnología de Alimentos (IATA), CSIC

Aim:

Using ozonation combined with aqueous maceration to isolate lignin fractions and derivatives from two different biomasses - *Miscanthus giganteus* (grass type), and *Vitis vinifera* (wood type) – that possess bioactive properties, including antioxidant, antimicrobial, UV absorption, and antifungal activity, for potential applications in food and food packaging.

Method:

A 200 L pilot-scale reactor was used for ozonation of *Miscanthus giganteus* and *Vitis vinifera* (vine shoot pruning residues). Samples were collected at different ozonation times, and ozone reaction products were extracted from biomass using water at 60°C. Physicochemical properties of the extracted fractions, including yield, lignin, molecular weight, and thermal stability, were analyzed. Additionally, the bioactive properties of the extracted fractions, such as antioxidant activity, total polyphenols, b-carotene bleaching, antimicrobial, antifungal activity, and UV absorption, were assessed.

Results:

Ozonation, as an oxidative process, effectively cleaved lignin from both biomasses, generating lower molecular weight lignin, phenolic monomers, and organic acids with bioactive properties. The efficiency of ozone depended on the initial lignin structure and its recalcitrance within the biomass. *Miscanthus*, as a grass type biomass, released lower molecular weight lignin and phenolic monomers, whereas vine shoots, as a woody biomass, released higher molecular weight lignin and organic acids. The difference in the composition of extracted fractions contributed to their bioactive properties. *Miscanthus* extracts exhibited higher antioxidant activity than vine shoots extracts. By extending the ozonation time, the antioxidant activity of the extracts decreased for both biomasses, but the extraction yields increased significantly by 5 to 7 times. More total polyphenols were extracted from both biomasses by extending the ozonation time. The extracts from *Miscanthus* demonstrated better antimicrobial activity against *Staphylococcus aureus* and *Escherichia coli* compared to those from vine shoots. Prolonging the ozonation time improved the antimicrobial activity of the extracted fractions from both biomasses. *Miscanthus* extracts exhibited better antifungal activity against *Botrytis cinerea* and *Penicillium digitatum* compared to vine shoots. All extracted fractions from both biomasses fully absorbed UVB and UVC, with vine shoots extracts showing superior UVA absorption.

Conclusion:

Ozonation is a green and efficient process to extract bioactive compounds from biomass with potential application in food and food packaging.

Tomato Leave Valorization in a Bioreactor for Enhanced Biomass Production

Ms. Duygu Mentés¹, Mrs. Ayse Sultan Akgun¹, Prof. Dr. Mecit Halil Oztop¹, Ms. Deniz Sevim Cabuk¹, Ms. Burcak Hakguden¹, Mr. Veli Baris Karatepe¹, Ms. Dilruba Hezer¹

¹Middle East Technical University

Aim:

This research investigates optimal conditions for efficiently converting tomato leaves (*Solanum lycopersicum*) into a valuable biomass source through hydrolysis and submerged fermentation. The research aims to identify a sustainable method for maximizing biomass yield from this significant agricultural waste product to produce single cell proteins.

Method:

Three different hydrolysis methods were applied to the tomato leaves: acid hydrolysis, alkali pretreated enzyme hydrolysis, and hydrothermally pretreated enzyme hydrolysis. The method yielding the highest concentration of reducing sugar and demonstrating greater sustainability was chosen as a fermentation hydrolysate. Next, fermentation was conducted with *Saccharomyces cerevisiae* in a 2L bioreactor under various parameters, including inoculation size (1, 3, 5% v/v), agitation speed (100, 200, 300 rpm) and fermentation duration (24, 72, 120 days). Initial and final reducing sugar concentrations were determined using the DNS method, while optical density at 600 nm was monitored throughout the fermentation process. Biomass obtained from fermentation will be further processed for single-cell protein extraction.

Results:

The results show that the highest reducing sugar concentration (23.91 g/L) was reached from 10% solid load by alkali pretreatment (1% NaOH, 15 min) followed by enzymatic hydrolysis with cellulase and hemicellulose. The preliminary fermentation results revealed significant biomass growth (OD₆₀₀ of 2.23) within 27 hours, utilizing 75% of the initial reducing sugar concentration.

Conclusion:

This research indicates the potential of utilizing tomato leaves as a sustainable carbon source for biomass production through optimized hydrolysis and fermentation. Maximizing biomass yield offers valuable opportunities for various downstream applications, including production of microbial proteins.

Water-based deep eutectic solvents as green method for the valorisation of spent coffee grounds

Francesca Pompei¹, Cinzia Mannozi¹, Matteo Tiecco¹, Sauro Vittori¹

¹Chemistry interdisciplinary Project (ChIP), School of Pharmacy, University Of Camerino

Aim: This research aims to extract bioactive compounds from spent coffee grounds using a green methodology focused on the application of water-based deep eutectic solvents (DESs). Spent coffee grounds, the residue obtained during the brewing process, contain several substances such as polysaccharides, alkaloids (e.g., caffeine) and phenolics compounds. While various studies have explored the extraction of bioactive substances using conventional DESs, such as choline chloride- or betaine-based solvents, the utilization of aqueous DESs, binary mixtures with water as a component, remains an open and unexplored field, also due to the novelty of these liquids. One of the aims of this work is the comparison of the results observed with *aquo*DESs with common volatile hydroalcoholic solvents in the extraction processes.

Method: The spent ground coffee purchased from local shops was air-dried and extracted with and ultrasound methodology. The different aqueous DES (glycolic acid:water, betaine:water, choline chloride:water) were compared to the conventional DES ethylene glycol:choline chloride and to the conventional hydroalcoholic solvent ethanol 70% and water. Then for each extracts the amount of phenolic compounds was defined according to spectrophotometric assays.

Results: The preliminary results are really promising, because the aqueous DES glycolic acid:water extracted a higher amount of phenolic compounds compared to the other aqueous DESs and also compared to ethanol/water. Moreover it was found to extract more efficaciously than water and ethanol/water. Ethylene glycol:water, in fact, extracted twice than water and 10% more than ethanol/water.

Conclusion: In a perspective of sustainability and recovery of food waste, employing water-based deep eutectic solvents represents a concrete innovation for the food industry. The extracts with those innovative green solvents, that possess antioxidant properties, could significantly decrease the impact of the waste generation and the usage of environmental harmful organic solvents.

Methodology Development for the Selection of Application-Specific E-Nose Calibrants: Focus on the Frying Process

Miss Lucia Corrà¹, Dr. Laura Capelli¹, Dr. Carmen Bax¹

¹Politecnico di Milano, Department of Chemistry, Materials and Chemical Engineering "Giulio Natta"

Aim:

In recent years, there has been a growing interest in the development of "smart" products, driven by continuous advances in industry digitalization, a cornerstone of the industry 4.0 concept (1). The electronic nose (E-nose), which consists of an array of low-cost gas sensors and a pattern recognition unit, is an excellent technology for developing "smart" objects with diverse applications. In the food sector, for instance, it can be used for process monitoring, determining food quality, and assessing sensory properties. (2). The development of application-specific E-nose devices require the execution of in-depth feasibility studies aimed at defining the most suitable hardware and software for the application (3). This paper proposes a unified methodology for the selection of application-specific calibrants that can be used for the execution of feasibility studies to perform a preliminary evaluation of the device's performance. Specifically, this methodology is exemplified by the development of a device to detect odour nuisances in the environment during frying and to monitor the process by analysing the volatile organic compounds (VOCs) generated.

Method:

A comprehensive literature search, including papers investigating the chemical composition of VOCs generated during the frying process, was carried out to identify the most representative VOCs for the specific application (4,5). The literature information obtained was organized in a table summarizing, for each reference, the main goal, the methodology for volatile sampling, the VOCs identified, and their concentrations. Subsequently, the application-specific calibrants were chosen based on the following criteria:

- Concentration: The most abundant compounds were preferred.
- Odour properties: The compounds with the greatest impact on the frying odour fingerprint (highest Odour Activity Values) were preferred.
- Chemical family: At least one compound from each chemical family were selected.

Results:

A great variability in terms of concentration was found in the literature due to the different objectives of the studies: e.g., quality of exhaust oil, odour nuisance due to frying, and organoleptic properties of fried food. When including all the variables, a total of 66 VOCs were identified: 26 aldehydes, 5 ketones, 9 nitrogen-containing volatiles, 3 hydrocarbons, 8 acids, 3 oxygen-containing heterocycles, 9 alcohols, and 3 sulphur-containing volatiles. 10 VOCs in total were selected: 2 aldehydes ((E, E)-2,4-Decadienal, Nonanal), 1 ketone (3-Nonen-2-one), 1 nitrogen-containing compound (3-Ethyl-2,5-dimethylpyrazine), 1 hydrocarbon (3-ethyl-2-methyl-1,3-Hexadiene), 1 acid (n-Hexadecanoic acid), 1 oxygen-containing heterocycle (2-Pentylfuran), 1 alcohol (1-Octanol), and 1 sulphur-containing compound (2,4-Dimethylthiophene).

Conclusion:

From this preliminary work, we were able to define a methodology for the selection of calibrants that is application-specific focusing on the monitoring of the frying process. Assessment of the reliability of the methodology will be performed with real-case scenario testing.

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Use of ultrasound and Vis-NIR spectroscopy to estimate moisture content in lasagna sheets

Dr. Anabella Giacomozzi¹, Marina Lopez Chulia², Jose Blasco², Jose Vicente Garcia Perez¹, Jose Benedito¹

¹Instituto de Ingeniería de Alimentos- FoodUPV. Universitat Politècnica De Valencia, ²Centro de Agroingeniería, Instituto Valenciano de Investigaciones Agrarias (IVIA)

Aim:

Water content is a critical parameter in the food industry, affecting product quality, safety, and shelf life. In the context of food analysis, visible and near-infrared spectroscopy (Vis-NIR) is a non-destructive analytical technique widely used due to its rapidity, simplicity, and ability to analyze multiple constituents simultaneously without the need for sample preparation. On the other hand, contactless ultrasound (US) uses air-coupled acoustic waves to perform fast and reliable non-destructive inspection of materials. This work aimed to explore the feasibility of using US and NIR techniques to estimate water content changes in lasagna sheets.

Method:

Lasagna samples were stored at different relative humidities for 14 days at 30 °C to induce changes in their moisture content, resulting in samples with moisture contents of 3.3, 8.6, 10.6, 12.5, and 17.6 % (w.b.). Ultrasonic measurements were taken at five points of the samples using contactless ultrasound sensors (0.28 MHz) in through-transmission mode. Signals were processed by calculating two ultrasonic parameters in the time domain (amplitude and velocity), to build a multiple linear regression (MLR) model. In addition, Vis-NIR spectral data were collected in reflectance mode using a multi-channel spectrometer equipped with three detectors covering the range 450 to 1700 nm. Each spectrum was obtained as the average of six scans at different points of the sample surface, covering both sides of the lasagna sheets. The Support Vector Machine (SVM) algorithm was employed to classify samples according to their moisture content. The dataset was divided by 80% and 20% for training and validation, respectively.

Results:

A close agreement between the experimental and predicted water content ($R^2=90.4\%$) using ultrasonic parameters was found. Moreover, the SVM model achieved 100 % classification accuracy when the lasagna sheets were classified according to their water content (5 groups). These results demonstrate successful, non-destructive, and rapid prediction of water content in lasagna pasta using both US and Vis-NIR technologies.

Conclusion:

Overall, Vis-NIR with contactless US offers a synergistic approach for predicting water content in lasagna pasta. However, further research is required to enhance the robustness of models to apply this combined approach at an industrial level.

Assessment of turbidity in beverage emulsions using computer vision

Arthur Gossen¹, Linda Katsch¹, Jan Schneider¹

¹OWL University of Applied Sciences and Arts

Aim:

The turbidity of beverages can affect consumer acceptance and can therefore be considered as a visual quality attribute. In emulsion-based beverages, turbidity is due to finely dispersed oil droplets. In fresh beverages, these oil droplets are homogeneously distributed throughout the bottle. However, over the course of ageing, various instability mechanisms can cause the oil droplets to grow, leading to creaming and ringing. In order to monitor this process, it was evaluated whether camera technology in combination with computer vision is suitable for quality monitoring and whether a corresponding monitoring device can monitor the turbidity of a bottle during storage.

Method:

For the computer vision model, images of model beverages and stored emulsion-based orange sodas are taken in a photo chamber. It consists of a darkened chamber, a light source positioned at 90° to the bottle, and a Raspberry Pi high quality camera with 16 mm telephoto lens. The model drinks are produced in such a way that the oil droplets cream as quickly as possible. Pre-processing steps for the model included removing reflections, cutting to a region of interest and converting pixels to greyscale. A random forest regression is then performed. As a reference, turbidity values in NTU are measured in the laboratory.

Results:

The determination of turbidity of model beverages is possible in the range of 0 to 350 NTU ($R^2=0.97$; RMSE=20 NTU). The determination of turbidity of stored orange sodas shows a reduced performance due to scatter in the range of 200 NTU ($R^2=0.82$; RMSE=11.1 NTU). Additional optical changes during the aging process of orange soda, in addition to turbidity reduction, could adversely affect the computer vision model. In addition to the image processing model, a beverage crate was built, incorporating the camera technology and an evaluation unit. This monitoring device can be used to assess the turbidity of beverages during storage.

Conclusion:

The results indicate the feasibility of using camera technology for measuring beverage turbidity. It can be performed effortlessly directly in bottles without sample preparation, making it suitable for a monitoring device. For final deployment, it is necessary to reduce model variability by incorporating uniformly distributed training data.

Towards targeted hydrolysis of Brewer's Spent Grain protein: Extraction, characterization, and prediction

Dr. Simon Gregersen Echers¹, Mr. Rasmus Kranold Mikkelsen², Dr. Lucas Sales Queiroz², Mr. Naim Abdul Khalek¹, Ms. Ioanna Fragaki², Ms. Panagiota Kolitsida, Dr. Betül Yesiltas², Dr. Timothy John Holey², Dr. Michael Toft Overgaard¹, Dr. Charlotte Jacobsen²

¹Aalborg University, ²Technical University of Denmark

Aim:

In this work, we aim to valorize brewer's spent grain (BSG) from beer production by application of emerging green technologies for protein extraction. Extracts serve as substrates in bioinformatics-assisted enzymatic proteolysis to produce hydrolysates with bioactive and/or functional properties for application as functional food ingredients.

Methods:

BSG proteins were obtained using a range of green extraction methods including pulsed electric field (PEF), microwave (MW), ultrasound (US), and Ohmic heating (OH) at different pH. Protein content and yields were evaluated using elemental analysis and mass balances and compared to conventional aqueous extraction. Together with unprocessed barley, malted barley, and raw BSG, extracts were quantitatively characterized for protein composition using bottom-up proteomics by LC-MS/MS. From this, bioinformatic algorithms were applied to predict potential emulsifier and antioxidant peptides embedded in abundant proteins. Synthetic versions of these peptides were obtained to confirm their properties in *in vitro* assays.

Results:

Overall, we found significant effects of the pH used during extraction on both protein yields and content, with a slightly alkaline pH being more efficient. Heating during extraction was found to generally improve protein extractability and the use of US resulted in a higher protein content. Moreover, significant protein-level differences were observed from barley compared to malt and BSG, but also observed between extraction methods.

We selected close to 100 previously uncharacterized peptides with predicted emulsifying or antioxidant properties. During evaluation, we found that a substantial number of these did indeed exhibit the predicted functionality. For instance, more than half of 48 assayed peptides effectively decreased oil/water interfacial tension and six of these even more so than caseinate. The peptides produced stable emulsions (5% oil in water) with very small droplet sizes at 0.2 wt% during storage.

Conclusions:

Green extraction technologies show promise for valorization of BSG by extraction and enzymatic hydrolysis of protein. *In vitro* and *in silico* data not only facilitates evaluation of extraction method suitability for obtaining high yields of proteins with highly functional peptides embedded in them, but also for designing targeted hydrolysis strategies, alleviating labor-intensive trial-and-error laboratory work.

Smart Odor Assessment of the Aroma Properties of Plant Proteins for Sustainable High Quality Products

Helen Haug^{1,2}, Finn Ehlerding³, Andreas T. Grasskamp¹, Andrea Bauer³, Vera Schoenhofen^{1,4}, Ute Schweiggert-Weisz^{1,5}, Gina Zeh¹, Tilman Sauerwald¹

¹Fraunhofer Institute for Process Engineering and Packaging (IVV), ²Friedrich-Alexander-Universität Erlangen-Nürnberg, Dept. Chemistry and Pharmacy, Chair of Aroma and Smell Research, ³Hamburg University of Applied Sciences, Life Sciences, Dept. Food Science and Nutrition, ⁴University of Bonn, Institute for Nutritional and Food Science, ⁵Technical University of Munich, School of Life Sciences, Plant Proteins and Nutrition

Aim: A changing product market and the strive for more sustainable, high quality foods make it necessary to develop efficient methods to assess and monitor product quality. Thereby, the sensory quality plays a major role, e.g. in alternative plant-based protein ingredients. We investigated the efficient evaluation of aroma features of plant protein derived from pea using the Smart Odor Assessment (SOdA) concept including the efficient collection and correlation of analytical and sensory data. The concept aims to enable the assessment of aroma characteristics based on analytical data in the future.

Method: Different pea protein samples were investigated in aqueous suspensions for human sensory assessments. The Rate-All-That-Apply (RATA) methodology was applied with 12 trained panellists using 31 sensory attributes. In parallel, pea protein samples were investigated instrumentally. Potent aroma compounds of a representative pea protein sample were identified using gas chromatography-olfactometry (GC-O) and multidimensional gas chromatography-mass spectrometry/olfactometry (GC-MS/O) and compared to literature data to evaluate potential aroma marker compounds. Headspace solid-phase-microextraction (HS-SPME) GC-MS was applied to investigate further samples. Analytical and sensory data was processed and statistical methods (e.g., partial least squares regression, and discriminant-based classification) were used for data correlation. The potential of this concept to assess aroma features using analytical data only was investigated.

Results: The sensory evaluation of the samples revealed that 20 of the 31 attributes significantly differed between the samples. In the representative pea protein sample, 15 potent aroma compounds were identified. HS-SPME GC-MS analysis allowed for the investigation of pea protein samples and detected several aroma compounds, including aroma compounds typically found in pea protein, as were supported by compound identification of the representative pea protein and comparison to data from the literature. Statistical methods indicated a high potential for classification and correlation of the analytical and human sensory data of pea protein.

Conclusion: The odor characteristics of different products can efficiently be determined by means of the SOdA-concept, as demonstrated using pea protein as an example. The concept can be adapted to other applications, e.g., other plant proteins or food raw materials. Funding: BMBF NewFoodSystem Innovation Space (031B0956P); Campus of the Senses (StMWi).

Low Cost MOS Gas Sensor Systems for Food Spoilage Monitoring

Mr Luigi Masi¹, Mr Dennis Arendes¹, Mr Johannes Amann¹, Mr Oliver Brieger¹, Professor Andreas Schütze¹, Dr Christian Bur¹

¹Saarland University

Aim:

Premature food waste can be avoided by food monitoring systems. For the assessment of food state, metal oxide semiconductor (MOS) gas sensors emerge as promising candidates due to their cost-effectiveness and ability to provide real-time in situ measurements. For these reasons, the aim of this work is to monitor the substances emitted by raspberries during the spoiling process by using calibrated MOS gas sensors.

Method:

To assess the gases that can commonly be found during the spoiling process of raspberries, a calibration of the sensors was performed in a gas mixing apparatus (GMA). The calibration of the sensors was performed with unique randomized gas mixtures made up with different quantities of eleven substances. These substances are selected based on literature studies of the common volatile organic compounds (VOC) detected during fruits' emission. Moreover, the employment of temperature cycled operation on these sensors was utilized to enable a more selective measurement of these gases while also expanding the concentrations' ranges of detectable substances. Finally, statistical models were built to estimate the changes of VOC during the spoiling process. Different amounts of raspberries along with gas sensors have been placed inside several closed food boxes allowing the sampling of the headspace of each box individually.

Results:

For each sensor and each gas, a regression model was developed based on the calibration of the unique gas mixtures. Subsequently, the validated model was used to estimate the VOC concentrations of the raspberries. The models have been evaluated in terms of root mean square error over the concentrations of the substances which composed the unique gas mixtures. Based on this, the built models then estimated the responses of the sensors to the raspberries over the days of experiment.

Conclusion:

Even though the absolute concentration has to be verified by means of analytics like GC-MS, the trend provided by the models reflect the observed behavior of the raspberries during the spoiling process. MOS gas sensors are therefore shown to be a promising solution to investigate the state of the food during the spoiling process.

Fiber Optic Sensor for Detection of Table Wines Using Artificial Intelligence Techniques

Fiber Optic Sensor For Detection Of Table Wines Using Artificial Intelligence Techniques jesus

Parada¹, Roberto Rojas-Laguna¹, Juan C. Hernández-García¹, Raul E. Sanchez-Yañez¹, Luis Granados-Zambrano¹, Maria E. Sosa-Morales¹, Maria S. Avila-Garcia¹, Marco Bianchetti¹, Stefano Toffanin²

¹Universidad De Guanajuato, ²Institute of Nanostructured Materials, Italian National Research Council

Fiber Optic Sensor for Detection of Table Wines Using Artificial Intelligence Techniques

Aim

Guanajuato is a leading wine producer in Mexico, home to over 12 internationally recognized vineyards. Certifying the authenticity of Guanajuato wines is crucial to protect local producers. This work aims to design a wine classifier using a fiber optic sensor and Support Vector Machine (SVM) techniques, achieving an accuracy greater than 97%. This classifier will detect table wines produced in Guanajuato, Mexico, ensuring their authenticity and safeguarding the state's wine industry reputation.

Method

A fiber optic sensor using the cone technique captured the absorption spectra of the wines displayed on an optical spectrum analyzer (OSA). Relevant features such as maximum peaks, wavelength, area under the curve and standard deviation were extracted using pattern recognition techniques. A database of 250 samples was generated with 5 labels, 4 labels corresponding to the mentioned characteristics and one to the type of wine. The k-fold cross-validation method split the data for testing and training, iteratively. Linear, Gaussian, polynomial and RBF SVM classifiers were trained and evaluated. Evaluation metrics such as precision, recall, F1 score, and specificity were calculated, and confusion matrices were displayed for each classifier. The Relief method determined the most important characteristics.

Results

Using the absorption spectra from our optical sensor, a classification model was developed. The objective was to achieve over 97% accuracy in identifying wine types. Four SVM methods were trained and evaluated. The results demonstrated an average accuracy of around 99.286%, significantly exceeding the objective. The polynomial SVM model stood out for its superior performance and low training time of 0.2503 seconds, making it the most promising method for classifying the analyzed wines.

Conclusion

This study demonstrated the effectiveness of Support Vector Machine (SVM) models for accurate classification of Guanajuato table wines based on their absorption spectra. The polynomial SVM model achieved an average accuracy of approximately 99.286%, surpassing the 97% goal, with a low training time of 0.2503 seconds. These results lay the foundation for reliable wine authentication tools, contributing to the quality and reputation of Guanajuato's wine industry.

Validation of On-Chip qPCR Process for *Staphylococcus aureus* Detection and Quantification: Methodological Overview

Msc Ana Pereira¹, PhD Alexander Oosterveld¹, PhD Gabriela Vollet Marson¹

¹One Planet Research Center

Aim:

On-chip quantitative polymerase chain reaction (qPCR) offers a new set of possibilities in the context of detection of pathogens in low volumes of sample, providing a fast and accurate quantification. This study presents a comprehensive validation framework for transitioning a qPCR process for *Staphylococcus aureus* from conventional benchtop equipment to on-chip technology.

Method:

Transitioning to on-chip qPCR introduces unique challenges, including managing reaction volumes, adapting to the chip's surface chemical properties affecting mixes, and navigating complexities inherent to miniaturized platforms. Overcoming these challenges is essential to ensure the reliability, accuracy, and efficiency of *S. aureus* detection and quantification using on-chip qPCR. Our strategy addresses four key aspects: applicability, specificity, sensitivity, and robustness. First, we evaluate the method's applicability by testing various matrices and DNA amounts to determine its scope. Rigorous testing, including different matrices and DNA concentrations, aims to identify potential qPCR inhibitors and understand method applicability.

Results:

Results demonstrate consistency across matrices while highlighting any identified interferences or limitations. Secondly, we assess method specificity by testing oligonucleotide sets against non-target and target materials. Experimental testing against reference materials ensures exclusive response to the characteristic analyte, minimizing false results. Thirdly, we establish the sensitivity of on-chip qPCR through a limit of quantification (LOQ). Utilizing standard curve and dilution series, we identify the minimal detectable analyte concentration., method robustness is evaluated by varying experimental conditions like primer and DNA concentration. Assessing resilience to changes ensures consistent results.

Conclusion:

In conclusion, our validation approach systematically addresses challenges to ensure the reliability and effectiveness of on-chip qPCR for *S. aureus* detection and quantification, facilitating its integration into diagnostic and food safety applications.

Identification of dietary fibers using broadband reflectance spectroscopy.

Msc. Claudia Tricanji¹, Steven van den Berg¹, Lien Smeesters²

¹The Hague University of Applied Sciences, ²Vrije Universiteit Brussel

Aim: This study aims to develop a non-destructive technology able to identify dietary fibers by combining ultraviolet-visible (UV-VIS) and near-infrared (NIR) diffuse reflection spectroscopy, covering the 350 nm to 1700 nm wavelength range. Specifically, a classification model was pursued differentiating the dietary fibers, while linking their optical properties to their chemical structures. As key contribution, this study intends to pave the way towards a fast, low-cost and environmentally-friendly method for measuring dietary fibers in food products.

Method: The measurements were carried out using a UV-VIS/NIR spectroscopy configuration comprising a reflection integrating sphere to collect all reflected light. The samples were illuminated by a deuterium and halogen light source (200 – 2500 nm). The collected light was subsequently analyzed by a broadband spectrum analyzer (Avantes AvaSpec) equipped with two detection channels for UV/VIS, and NIR light respectively. Spectral data were processed by calculating wavelength ratios to discern the unique molecular "fingerprints" of each fiber type—marked by specific combinations of molecular features of each dietary fiber. Dietary fibers analyzed include Physilium husk powder, Inulin powder branded as Frutalose and Oligofruktose/Frutafit Inulin, high methylation (HM) Pectin powder, and two types of Cellulose powder from Sigma Aldrich and Acros Organics.

Results: Most significant spectral features were identified in the NIR range, while in the UV-VIS channel the differences were observed with lower intensity. The spectral data from both channels were processed using a wavelength ratio algorithm, resulting in effective data clustering of the dietary fibers' spectral signals. Inulin and pectin showed to be clearly identifiable. Cellulose and Physilium displayed very similar characteristics.

Conclusion: This study demonstrates that UV-VIS and NIR spectroscopy in combination with wavelength ratio calculations is a promising method for dietary fiber analysis. The distinct clustering of fibers, indicative of their chemical composition, highlights the method's effectiveness. Future work will focus on applying this methodology to a broader spectrum of dietary fibers and developing calibration models for determining fiber matrices in food products. This advancement could contribute to enhanced nutritional labeling and dietary recommendations.

Development of automated food quality monitoring systems based on low cost sensors

Maximilian Koehne^{1,2}, **Thorsten Tybussek**¹, Tilman Sauerwald^{1,2}, Helen Haug¹, Gina Zeh¹

¹Fraunhofer IVV, ²Saarland University

Aim:

The quality of oils and fats is highly dependent on the degree of oxidation. For the determination of the degree of oxidation, mostly chemical approaches are used, which are time consuming and require a laboratory infrastructure. The aim of this work is therefore to develop a general concept for GC systems based on low-cost metal oxide semiconductor (MOS) sensors for specific requirements, such as the monitoring of nut quality using oxidation markers.

Methods:

To determine the degree of oxidation of the tested food products, the peroxide value or the anisidine value was determined in a chemical approach. By combining both, the total oxidation value (TOTOX-value) can be calculated, which is used as ground truth. To identify relevant volatile compounds in the headspace, several samples were analyzed by GC-mass spectrometry (MS). In addition, the GC-selective odorant measurement by sensor array (SOMSA) method was used to shorten time-consuming analyses for characterizing sensor responses.

Results:

Within the SOMSA approach, the MS was able to detect the individual oxidation markers. A comparison with the sensor signal response also showed that individual sensor layers of the four-layer sensor used here are particularly suitable for individual markers. Therefore, it may be possible to save an upstream separation unit in the form of a GC column for future GC sensor systems.

Conclusion:

The rapid identification of individual relevant markers and the parallel characterization of the sensors eliminates the need for time-consuming laboratory analysis, thus enabling the SOMSA method to save time and money as part of the development platform for GC sensor systems. Using nuts as an example, it was possible to characterize rancidity as a quality marker on a sensor.

Development of a visual inspection tool for quality-based management of CA storages

Sander Van Wayenbergh¹, Prof. Bart Nicolai¹, Pieter Verboven¹

¹Ku Leuven

Aim:

Quality classification of fruit after harvest is today still manual and subjective based on sampling by experienced inspectors. The objective of this work was the development of a software tool supported by artificial intelligence (AI) for large-scale data collection on the color and size of pome fruits (i.e. apples and pears) in industrial size bins. The tool should operate in a fully automated manner to minimize the effort spent on sampling since the time between fruit harvest and incubation in ultra-low oxygen (ULO) storage rooms is limited. The obtained information should allow to better match the quality of fruit in a storage room to the demand of the fresh fruit market.

Method:

Data collection in this case relies on RGB image processing. A pipeline was developed to enable processing of images taken with mobile phones or with a fixed camera set-up using color references for image calibration. Fruit in the input images are first segmented by image segmentation algorithms prior to fruit size estimation. In the mobile case, perspective correction methods are applied to correct for the angle under which the picture was taken. The focus for AI development lies on robustness in different environmental conditions and easy adaptation to multiple cultivars and fruit species.

Results:

The image processing pipeline can process images of fruit bins in a fast and accurate manner for size and color detection of pome fruits. With an accuracy of 2-3 mm, the majority of fruits are subdivided in their correct size classes. Meanwhile, the current run time of 10s per image allows to keep up with the operational flow in storage facilities. Initial results on the robustness of the algorithms to different lighting conditions seem to indicate that the need for a controlled environment is low. However, further testing in outdoor environments with varying weather conditions is needed to guarantee robustness.

Conclusion:

The developed software tool is capable of extracting accurate information about the size and color from an industrial size fruit bin containing apples or pears. This offers opportunities for quality assessment at every point along the postharvest cold chain and thereby reduces the need for manual measurements.

Raman spectroscopy as a tool for investigating secondary structure of proteins in food systems

MSc. Mathijs van Kilsdonk¹, MSc. Ana J. F. Q. R. Pereira¹, PhD. Alexander Oosterveld¹, Phd Gabriela Vollet Marson¹

¹OnePlanet Research Center

Aim:

Proteins are complex molecules playing essential roles in the body and in food systems. Protein functions (eg., enzyme activity, gelation, foaming) may be altered depending on the extent of degradation that these molecules undergo during processing. Information on the proteins' secondary structure give insights into the denaturation level or enable detection of specific proteins in a sample. In this study we propose the use of Shifted-Excitation Raman Differential Spectroscopy (SERDS) as a non-destructive tool to assess the secondary structure of proteins in solution. The Raman fingerprint is subsequently used to determine the presence of protein fractions in solutions.

Method:

For SERDS measurements of the protein solutions, spectra were collected for two lasers with center wavelengths of 784.4 and 785.5 nm, using sequential illumination. Prior to statistical analysis and interpretation, the Raman spectra were reconstructed using linear operations. To evaluate SERDS' ability to discern varying protein fractions, a mixture series of 18 samples with changing proportions of alpha-lactalbumin and beta-lactoglobulin was set up. With the intent of mimicking the whey matrix, the total protein concentration was maintained at 5 g L⁻¹, with a constant lactose background of 42 g L⁻¹. The reconstructed Raman spectra were correlated to the protein fraction via partial least squares regression and leave-one-out cross validation. Recursive feature selection was implemented to improve the performance.

Results:

SERDS accurately distinguished protein solutions with varying proportions of the two proteins. Predictions ranged from pure alpha-lactalbumin to pure beta-lactoglobulin, with a root mean square error of 4.8% in alpha-lactalbumin fraction and a strong linearity ($R^2 = 0.96$). Recursive feature selection enhanced performance, with selected spectral features corresponding to protein secondary structure.

Conclusion:

In conclusion, SERDS emerges as an accurate method for assessing protein fractions in solution. Its capacity to differentiate between protein proportions with low error rates and high linearity shows its potential in biological and food systems. The correspondence between variable selection and spectral features related to secondary structure highlights that Raman spectroscopy is effective in assessing the structure of proteins.

Real-Time Detection of Turmeric Adulteration with Metanil Yellow using Miniaturized NIR Sensor and AI techniques

Msc Student/researcher Dimitra Xenitopoulou¹, Nikolaos Tsakiridis¹, Achilleas Zalidis¹, Eleni Kalopesa¹, George Zalidis^{1,2}

¹Spectra Lab Group, Laboratory of Remote Sensing, Spectroscopy, and GIS, Department of Agriculture, Aristotle University of Thessaloniki, ²Interbalkan Environment Center

Aim: Products like spices have been among the most targeted foods in the European Union for fraudsters, given that the spice market exemplifies complex and globalized supply chains. Turmeric is a widely used spice famous for its vivid color, unique flavor, and purported health advantages. Its medicinal properties in addressing various health issues have sparked a surge in global demand for this spice, raising concerns about the spice industry's integrity. The most common adulterants of turmeric, added for financial gain, are synthetic, non-authorized azo dyes, particularly Metanil Yellow (MY). To tackle eventual malpractices concerning the turmeric supply chain, this study addresses the exploration of a rapid, cost-efficient, and non-destructive method utilizing a miniaturized NIR (1350 – 2500 nm) sensor coupled with Artificial Intelligence (AI) techniques to detect the presence of MY in turmeric.

Method: A miniaturized NIR sensor recorded the reflectance of 100 authentic and adulterated turmeric samples (various levels of adulteration: 5, 10, 20, 30, 40% w/w MY). AI regression algorithms were then trained to identify anomalies indicative of turmeric adulteration with MY, while examining multiple spectral pre-treatments like the first-derivative. To put the results into perspective and evaluate the accuracy of the low-cost miniaturized NIR sensor, its results were compared with a high-precision benchmark spectroradiometer, both in terms of spectral signal and model accuracy.

Results: The developed AI regression models demonstrated satisfactory accuracy in identifying adulteration of turmeric with MY (R^2 of 0.78 using the first-derivative spectra and the XGBoost model), thereby providing a reliable tool for real-time monitoring of turmeric authenticity. In contrast, the best model from the benchmark spectroradiometer had an accuracy of estimation R^2 0.84 using the first-derivative spectra. The mean distance between two spectral measurements of the same sample using spectral angle mapper was 0.08.

Conclusion: The proposed approach represents a promising non-destructive solution for on-site testing of turmeric authenticity at various supply chain stages, enhancing transparency and accountability in the spice industry and strengthening consumer trust. Moving one step further, integrating portable NIR sensors and AI modeling into a user-friendly platform potentially facilitates the widespread adoption of the developed technology.

Validation of an electronic nose sensor to monitor fruit and vegetable processing through aroma signature

Dr Alexandre Leca¹, Alejandro Villamarin¹, Christian Ginies¹, Dr David Page¹, Dr Sylvie Bureau¹

¹INRAE - UMR408 SQPOV

Aim:

Fruit and Vegetables industry faces the urgent need to reduce loss and waste and energy consumption, while maintaining pleasant organoleptic qualities. We tested a volatile compounds electronic sensor to evaluate its ability to detect various plant-based food products and changes in the aroma of apple puree products in humid food processing conditions. Such electronic nose sensor (E-nose) would provide a significant asset in fast monitoring of aroma signature, after a product validation and a supervised learning.

Method:

An E-nose (NeOse Advance, Aryballe, Grenoble, France) was used. Its principle relies in an array of 64 bi-branched Mach-Zender interferometers, each with a blank and a functionalized arm with peptides showing affinities for various volatile compounds. The NeOse having never been tested against fruit and vegetable products in humid conditions, and despite an embedded humidity sensor designed to correct the signal as a function of environmental changes, we had to quantify its limit of detection (LOD) of a reference molecule (6-methylhept-5-en-2-one) as well as that of apple puree. We also evaluated the variability of the measured signal for various plant purees (apple, carrot, citrus, leek, tomato) and different cooking stages of apple puree (raw, semi-cooked, cooked, overcooked).

Results:

The E-nose linearly detected the reference molecule up to a concentration of 2 ppm, i.e. significantly lower than the total volatile compound concentration in plant-based foods. As a confirmation of the E-nose's sensitivity to fruit & vegetables products, its LOD for apple purees was significantly below typical apple puree concentrations ($R^2 > 0.9$ for linear models at low dilution).

Principal Component Analyses were performed to evidence the E-nose sensitivity to: i) differentiate the specific aroma signature of lemon juice and leek, carrot, apple, tomato purees, (79% of variance explained by two principal components); ii) accurately discriminate raw, semi-cooked, cooked and overcooked apple purees by their aroma signal, mainly along a principal component explaining 58% of the total variance.

Conclusion:

This study evidenced the robustness of the E-nose to detect various food products as well as their change in aroma occurring during cooking. Further studies will test the robustness of the E-nose in real-time during food processing.

Use of X-Ray Microtomography for the Analysis of Plantain during Hot Air Drying

Dr. Ayobami Olayemi Oladejo¹, Sebastian Gruber¹, Michaela Thalhammer², Prof. Heiko Briesen², Prof. Petra Först¹

¹Food Process Engineering, TUM School of Life Sciences, Technical University of Munich, Weihenstephaner Berg 1, 85354, ²Process Systems Engineering, TUM School of Life Sciences, Technical University of Munich, Gregor-Mendel-Str. 4, 85354

Aim: X-ray microtomography (XCT) is a useful tool that can reveal the internal structure of a food product non-invasively during drying. The aim of this work is to quantify and monitor the drying behaviour (evolution of porosity, pore size, pore size distribution and shrinkage) of plantain chips during hot air drying using XCT in order to optimize the mechanical and textural properties of the product.

Method: Plantain was sliced into diameter and thickness of 5 and 3mm, respectively. The plantain chips were subjected to hot air drying at temperature of 50, 60 and 70°C and air velocity of 1m/s. The microstructure of fresh samples before and during drying was determined by XCT operating at 80KV and 50µA. Scans were made with a resolution of 4µm in voxel size. The 2D radiographs obtained were reconstructed to 3D images for further analysis. During drying, the plantain sample was taken out of the dryer at intervals of 30 min for mass and XCT measurements until constant mass was attained.

Results: The result showed that there was an initial increase in the porosity of plantain samples till a moisture ratio of 0.6 was attained and thereafter the porosity decreased as drying progressed. Furthermore, higher temperature led to higher porosities in the dried samples. The pore size of the samples increased as the moisture content decreased during drying. At 70°C, the pore size distribution shifted towards larger pore sizes as drying progressed. The shrinkage ratio showed that there was 15, 20 and 22% volume reduction for samples dried at 50, 60 and 70°C, respectively, at the end of drying. XCT also revealed the growth of crust formation during drying at 70°C.

Conclusion: Consequently, this work showed that high temperature strongly influenced the porosity, pore size distribution and shrinkage of plantain, while moisture loss influenced the formation of pore size during hot air drying. The results obtained through XCT could help in modelling, prediction and optimizing of process parameters during hot air drying of plantain.

Evaluation of drift potential of hollow cone nozzles on airblast sprayer by novel resistive method

Ms Ayesha Ali¹, Mr Lorenzo Becce², Mr Antonio Altana³, Prof Fabrizio Mazzetto^{1,2,3}

¹Faculty of Agricultural, Environmental, and Food Sciences, Free University of Bozen-Bolzano,

²Competence Centre for Plant Health, Free University of Bozen-Bolzano, ³Competence Centre for Mountain Innovation Ecosystems, Free University of Bozen-Bolzano

Aim:

Drift is one of the major challenges associated with the application of plant protection products. It is of utmost significance to assess the drift for resource management and to reduce the off-target material deposition and its associated negative impact on bystanders and the environment. The commonly used methods for drift assessment are time-consuming and lack real-time estimation of the deposition. This calls for new methods that could enable the real-time estimation of the drift.

Method:

The presented study proposes a resistive-based method that can be used as an alternative to optical methods to quantify the drift potential of spraying equipment in real time to enable better control and quick actions. The drift potential of an airblast sprayer fitted with hollow cone nozzles was evaluated in the wind tunnel in the Agroforestry Innovations Laboratory (AFI-Lab) of the Free University of Bolzano. A tap water-based fluorescein solution, a low-cost tracer widely used in drift assessment studies, was sprayed by a commercial orchard airblast sprayer to mimic plant protection products. Two horizontal test benches equipped with sliding covers were placed parallel to each other. The first test bench remained open during the whole pass of the sprayer to collect ground deposits, while the other was revealed after spraying, to collect the spray fraction settling more slowly, which is more prone to drift. Silver screen printed electrodes were incorporated in the collectors and connected to a Sciospec impedance analyser (ISX-5v2) to measure the real-time impedance of the collected material. To ensure repeatability and reproducibility, three replicates were performed under comparable meteorological conditions (wind speed, temperature, and relative humidity).

Results:

To ensure the accuracy of the measurement, the results were compared with optical measurements performed simultaneously. The results of both methods were found comparable.

Conclusion:

In conclusion, the resistive-based approach yields promising results, enabling faster and reliable tests of spray drift potential under controlled and field conditions alike.

Computational Design of Curved-Shape Baffles in Shell and Tube Heat Exchangers

PhD Candidate Kubra Bulduk Sahin¹, Assistant Professor Mustafa Ozturk², Engineer MSc Murat Sahin³, Professor Ferruh Erdogdu¹

¹Ankara University, Department of Food Engineering, ²Sakarya University, Department of Food Engineering, ³Tanpera R&D Center

Aim:

Computational modelling of flow patterns on shell side of shell and tube heat exchangers is a challenging issue. Modifications in baffle geometry and configuration have a direct impact on the flow evolution. For evaluating a shell and tube heat exchanger's process efficiency, main factors to consider are the thermal efficiency and low pressure drop. Use of baffles to improve these factors is the general approach. Therefore, the objective of this study was to develop a computational model for designing an industrial scale shell and tube heat exchanger and to evaluate its efficiency with the use of curved-shape baffles.

Method:

Computational modelling studies were carried out using Solidworks Flow Simulation (V.2019), and the Bell-Delaware approach was employed to validate the developed models. Milk was used to represent a liquid food product on the tube side while superheated steam was utilised on the shell side. In the simulations, a conventional segmental baffle was employed to evaluate the performance of the curved-shape at various bending radii in two different baffle configurations according to its orientation in the shell side. Optimal design of the curved-shaped baffle radius was determined based on the evolved prediction model of overall heat transfer coefficient.

Results:

Following the model validation, while evaluating the results of baffle configurations, the baffle was positioned in a relative distance to the inlet nozzle on the shell side leading to the increase of thermal and hydraulic performances. The novel curved-shape baffle was also determined to outperform the conventional segmental baffle, and optimum curved-shape baffle achieved the highest effect at 670 mm with a temperature increase of 21°C and a pressure drop decrease of 2634 Pa compared to the segmental baffle.

Conclusion:

The result of this study highlighted that baffle orientation inside the shell had a direct impact on thermal and hydraulic efficiency. Furthermore, the application of a curved shape geometry baffle had a notable impact on its performance, enhancing the flow evolution and resulting in energy savings as compared to a segmental baffle.

Simulation of emissions from salmon post-harvest steps: comparing the ENOUGH tool with a LCA study

Hanne Dalsvåg¹, Lukas Köster¹, Graciela Alvarez²

¹SINTEF Ocean, ²L'Institut national de recherche pour l'agriculture, l'alimentation et l'environnement (INRAE)

Aim:

The global demand for food is increasing, intensifying pressure to food systems related to food quality and greenhouse gas (GHG) emissions in the whole food chain. In Norway, salmon aquaculture is an important food industry. The majority of produced salmon in Norway is exported, making the transport type and distance important sources of emissions. Environmental assessments of food are getting increased attention, and in 2020 a comprehensive life cycle assessment (LCA) report analysing GHG emissions from salmon products dominating the export statistics was published. In the ENOUGH project, a web-based simulation tool is developed, enabling simulation of the food chain of selected products, estimating GHG emissions along the chain to identify the most critical points where improvements can be applied. The aim of this work was to establish a comparison of the ENOUGH tool to the LCA report to potentially indicate the benefits and shortcomings of the tool.

Method:

Within this work, post-harvest cold chains for Norwegian salmon were simulated in the ENOUGH simulation tool and compared against a published life cycle assessment report on the Norwegian fisheries and aquaculture sector. Thereby, similarities and differences between LCA results and the ENOUGH tool outputs were investigated, and the advantage of a detailed cold chain simulation tool was explored.

Results:

The simulation output indicates that packaging and transport dominates the post-harvest emissions of the investigated salmon chain. The point of difference between the ENOUGH tool compared to the life cycle assessment report is the focus on the cold chain, with determination of temperature-time profiles of the specific food chain, and taking refrigeration specific details into account when calculating emissions. One advantage of the tool is its evaluation of food quality, which was not investigated in detail in this work.

Conclusion:

In conclusion, there are several ways to calculate emissions from food systems, yielding somewhat different results. Not necessarily being as detailed as a full LCA analysis, the investigated simulation tool can still provide user-friendly decision support to the food industry. Given the complexity of food systems, including quality calculations in combination with emissions could also be an advantage.

Smart control of apple puree processing based on key aroma compound evolution

Phd Ahmadou Moustapha Dieng¹, Dr Sylvie Bureau¹, Dr David Page¹, Dr Christian Ginies¹, Dr Raphaël Plasson², Dr Isabelle Souchon¹, Dr Alexandre Leca¹

¹Inrae, ²Avignon Université

Aim:

The underlying mechanisms of the physicochemical transformations occurring during apple puree processing are complex. These mechanisms limit a smart control of processing yet essential for variable raw materials in order to optimize energy consumption and minimize the loss and waste during processing. We thus studied key steps of apple puree processing, aiming at modelling the change in organoleptic properties over time. Alongside the various organoleptic quality variables we investigated, this study is focused on the aroma compounds evolution at successive key steps of the process.

Method:

All purees were made in a semi-closed cooker-cutter (RoboQbo, Bentivoglio, Italy) following a DoE of two target temperature levels (70, 95°C) and two grinding speed levels (300, 1000rpm). Sampling was conducted at four key steps: after 5 minutes at 25°C (T25), after 5 minutes at 45°C (T45), upon reaching the target temperature, and after 30 minutes cooking at the target temperature. Aroma analysis was performed by coupling a purge and trap system to a GC-MS (Trace1300 and ISQ LT, ThermoFisher Scientific, USA).

Results:

Among the 52 compounds identified, we selected those with the lower intra sample Mean-Relative-Error (<30%) and evolve over steps (increase or decrease) leading to a follow-up of 15 aroma compounds : acetaldehyde, propan-1-ol, butanal, 2-methylpropan-1-ol, propyl acetate, 2-methylbutan-1-ol, pentan-1-ol, n-hexanal, furfural, 2-methylbutyl acetate, pentyl acetate, 6-methylhept-5-en-2-one, hexyl acetate, (E)-2-hexenal and butyl propanoate. Their quantifications at each processing step show that the combination of the higher temperature (95°C) and the lower grinding speed (300 rpm) induces a greater impact on compound variation. The set temperatures 70°C and 82.5°C generally had a limited effect on aroma compound profile, except for the highest volatile or the most reactive compounds as acetaldehyde, propyl acetate, n-hexanal, (E)-2-hexenal and 6-methylhept-5-en-2-one. At T25 and T45 no difference in aroma composition were noted.

Conclusion:

Through this work we were able to selected 15 volatile compounds, released and/or consumed during apple puree processing. These will be implemented in a model to be validated on new apple puree processes. Then eventually be part of a predictive framework to manage apple puree processing conditions to reach a optimum quality.

AI-based surrogate models of digital twins for food and drink manufacturing systems

Emmanuel Lwele¹

¹Sheffield Hallam University

Aims and Objectives

Digital twins (DT) are virtual replicas of physical systems, products, or processes that enable simulation, optimisation, and prediction of their behaviour, with the help of Artificial Intelligence (AI) solutions applied to Industry 4.0.

The core aim of this project is to investigate novel machine-learning architectures for data-driven surrogate modelling in food and drink manufacturing systems. The main objectives needed to support this aim are:

1. Develop an Industry 4.0 exemplar system within NCFE to acquire real-world data from food processing machinery.
2. Develop a Digital Twin model of this system based on both system process dynamics and the integration of acquired sensor data.
3. Create computationally lightweight machine learning-based surrogate models combining data from the Digital Twin model and real-world sensor data to use in design space exploration and advanced model predictive control.

Methodology

The methodology and framework to be used for this project can be broken down into the following main stages:

- i. Identify a food and drink manufacturing exemplar process and create a test bench via instrumentation with Industry 4.0 sensors.
- ii. Develop a digital twin model capturing the process dynamics of the exemplar system (e.g., by integrating CFD/DEM/FEA modelling techniques - as appropriate - with real-world data).
- iii. Create lightweight computationally tractable ML/AI-based surrogate models from the complex digital twin models capable of being used in rapid design space exploration and advanced model predictive control strategies.
- iv. Validation and evaluation of ML/AI-based surrogate models built in (iii).

Results

The specific expected outcomes of this project are:

1. Deeper insight into the dynamic behaviour of the exemplar food and drink manufacturing process to allow efficient production designs.
2. A novel set of AI-based methods for creating data-driven surrogate models that are not only applicable to the food manufacturing industry but also to other manufacturing sectors.

Conclusion:

The coupling of advanced machine learning-based surrogate models and digital twin technology for design space exploration, anomaly detection, and model predictive control is a highly novel area - with applications not only to the food and drink manufacturing industry but to other manufacturing sectors.

DEVELOPMENT OF A DIGITAL TWIN IN THE PORK SECTOR

Mónica Mendiola Lanao¹, Rafael López¹, Jose María Alonso², Héctor Arias², Daniel Prieto², Daniel de la Puente¹

¹CNTA, ²SOLTEC ingenieros

Aim: The European project BBTWINS is dedicated to revolutionizing the agri-food value chain through the implementation of digital twins, with a particular focus on pork and cured ham production. This initiative leverages cutting-edge technologies such as artificial intelligence (AI), the Internet of Things (IoT), blockchain, and big data analytics to enhance and optimize every stage of the production process.

Method: In the value chain of pork production, digital twins serve as virtual counterparts to physical processes, enabling real-time simulation and analysis. IoT sensors deployed on farms gather extensive data on environmental conditions, as well as the health and behavior of pigs. This data is then transmitted to a digital platform, where AI algorithms process it to create predictive models and generate optimized recommendations. For instance, by analyzing data, the system can detect behavioral patterns that may indicate health issues, allowing for early interventions and thereby reducing mortality rates.

During the cured ham processing phase, digital twins facilitate meticulous control over curing conditions. Sensors monitoring temperature and humidity continuously collect data, which is used to automatically adjust curing parameters in real-time. This ensures consistent product quality and minimizes waste. Furthermore, the integration of blockchain technology into the platform ensures complete traceability of the product from the farm to the final consumer, thereby providing a transparent and secure supply chain.

Results: Big data analytics is pivotal in optimizing the entire value chain. The data collected throughout the process is analyzed to pinpoint areas for improvement and to develop strategies for optimization. For example, data analysis can uncover inefficiencies in resource usage, such as water and feed, enabling adjustments that enhance sustainability and reduce operational costs.

Conclusion: In summary, BBTWINS signifies a major leap forward in the digitalization of the pork and ham value chain. By offering innovative technical solutions, it aims to improve the efficiency, quality, and sustainability of meat production across Europe. This project not only enhances productivity but also ensures a higher standard of animal welfare and environmental stewardship, setting a new benchmark for the agri-food industry.

Mechanistic modelling and kinetic of plant-based meat alternatives

Ms. Anna Christodoulou¹, **Dr. Aberham Hailu Feyissa**¹

¹Food Production Engineering, National Food Institute, Technical University Of Denmark (DTU)

Aim:

Despite the rapid progress in plant-based meat alternative development, there remains a noticeable gap in research focused on deeper understanding the quality transformations that arise during further processing steps like thermal processing, frying or roasting. These processes involve complex phenomena such heat and mass transfer in porous matrix, dynamical material changes, resulted with process-induced quality changes. Therefore, the current study aims to develop a mathematical model of the transport processes coupled the kinetics of quality changes during the contact frying of plant-based meat analogues.

Method:

The mathematical model has been formulated from mechanistic understanding of plant-based meat analogues. The differential equations that describe the transfer phenomena and kinetic of quality changes were solved using COMSOL Multiphysics® 6.2, using the Finite Element Method and state variables and quality parameters were predicted. The experiments were carried out under different process conditions. To develop kinetic model, a new systematic experimental approach (at small scale) was developed to the link local state variables (e.g. temperature, moisture) to quality parameters (such as texture, colour, etc). The approach to map spatial (e.g., moisture content and temperature) and quality parameters. Temperature and moisture profiles, colour changes were monitored continuously throughout the frying process. Validation experiments were performed at large scale for all quality parameters at different process conditions.

Results:

A mechanistic model coupled kinetic models was established and validated using the experimental data and good agreement was obtained. Water release to the porous material was observed due to protein denaturation, thus the moisture transport is driven by the pressure gradient. Higher process temperatures lead to rapid textural and colour changes and leading losses of moisture. However, insignificant deformation of the product was observed.

Conclusion:

A mechanistic understanding of the driving phenomena is essential for effectively controlling and optimizing the products. The developed model enables deeper insights into the localized and spatial changes in quality parameters in plant-based meat analogues during the contact frying process. The model can be used for prediction, optimizing and as digital twin tool by further incorporate with sensors.

The brute force simulation of hydrothermal driven losses and compositional data analysis of phyco ingredients

Dr Thiruchenduran Somasundaram¹, Dr Thomas Mock¹, Dr Damien Callahan², Dr David Francis¹

¹Nutrition and Seafood Laboratory (NuSea.Lab), School of Life and Environmental Sciences, Deakin University, ²School of Life and Environmental Sciences, Deakin University, Burwood Campus

Aim: This study aims to employ brute force simulation and a marker nutrient based compositional data to estimate the losses incurred to *Phyllospora comosa* nutrients during a hydrothermal experiment.

Method: A reference nutrient based additive log ratio transformation alleviates the compositional constraints. A reference nutrient is selected based on its hydrothermal process stability. This paper describes two objective methods for the estimation of the hydrothermal stability of *Phyllospora comosa* crude nutrients. The unit proportion method calculates the total mass loss required by each nutrient to reach its final composition. A brute force simulation estimates the true retentions of each nutrient at their minimal loss.

Results: The lipid based additive log-ratio transformation and analysis resulted in large, significant ($P < 0.05$), and negative effects of the treatments on the nutrient contents with Pearson's product moment correlation of 83%, 96%, and 90% for protein, ash, carbohydrate, respectively. A 53% total mass loss was estimated during the hydrothermal process simulation.

Conclusion: The new methods described here in for the selection of a reference nutrient and the reference nutrient based additive log-ratio transformation and analysis showed that a compositional dataset can unravel true contentual behaviours of the nutrients during a hydrothermal processing. Slight modifications to the algorithms can help the selection of a reference biomarker in bioprocessing experiments and experiments that result in microbiome, metabolomics, and other compositional datasets to be used as a reference in the additive log-ratio transformations.

Computational Design of Microwave System Coupled with Air Impingement for Temperature Uniformity Enhancement

Kubra Polat¹, Huseyin Topcam¹, Ozan Karatas¹, Dr. Ferruh Erdogdu¹

¹Ankara University

Aim:

Microwave processing is an efficient novel green technology with its process efficiency. A major challenge in a microwave process is the formation of non-uniform electromagnetic field evolution with resulting uneven temperature distribution. This is significantly observed along the surface of the products. This study aims to enhance the surface temperature uniformity during microwave processing by incorporating an air impingement system into a lab-scale microwave system and computationally analysing the efficiency of the process.

Method:

A computational model was developed to determine the electromagnetic field evolution inside the microwave cavity and temperature distribution within the product during the air impingement-assisted microwave process. Comsol Multiphysics software (V. 6.2) was used for model development. For experimental studies, a cylindrical sample (2% agar in 9.9×1.8 cm) was placed within the cavity of a 2450 MHz system, and microwave heating was carried out. To enable air impingement, a funnel-shaped custom-printed head (with 15 nozzles – 7 mm diameter) coupled with a blower was inserted into the cavity through a hole at the cavity top surface to place 6.5 cm away from the top surface of the agar sample. Temperature measurement within the agar sample was carried out with fibre optic probes.

Results:

Computational model results for the impingement processing compared well with the experimental data obtained at two distinct points within the sample. Then, the temperature profile of the sample was compared with the results from the conventional case to demonstrate the effect of air impingement on the surface temperature evolution. Due to the limited air velocity at the experimental conditions (less than 3 m/s at nozzle exits), overall heating rate was not significantly reduced while a significant temperature uniformity along the sample surface was observed. Increased impingement air velocities were also applied for the microwave thawing process resulting in a uniform temperature evolution compared to the conventional processing.

Conclusion:

The results indicated an efficient use of the impingement process with microwave heating. Increased temperature uniformity specifically for the thawing process showed promise for this approach in an industrial setting where radio frequency heating is preferred with its longer process time.

Towards optimal seed drying: Modelling drying and its effect on seed quality

Julia Veser¹, Ruud van der Sman¹, Maarten Schutyser¹

¹Laboratory of Food Process Engineering, Wageningen University & Research

Aim:

High-quality seeds are the starting point for successful agricultural plant production and have an impact on the sustainability of our food production system. Seed breeding companies are challenged by producing high-quality seeds for farmers and keeping the quality of different batches constant to assure healthy plants and high yields. Therefore, throughout the seed production process, many dedicated treatments and drying steps are applied. Drying treatments have not yet been intensively studied. In particular, the specific effects of drying on storability and germination, as well as optimal drying routes are largely unknown. Therefore, we investigate drying of seeds by modelling the drying kinetics and linking them to predictions of quality.

Method:

We focus on a single seed level of simple shaped cabbage. For validation of the models, a small-scale convective air dryer was compiled in combination with in-line measurements and a humidifier to precisely control air conditions in terms of temperature and relative humidity (RH). After drying, the germination quality was tested with germination tests on paper. To enable optimisation, a mechanistic model was derived describing heat and mass transfer during drying of seeds, where diffusivity and moisture sorption were estimated via separate experiments and seed shrinkage was considered. In addition, response surface modelling was used to describe the germination quality data.

Results:

Varying drying conditions influenced final seed germination quality whereas higher temperatures above 55°C lead to decreased quality. Quality might be closely linked to reaching the glassy state, the cell membrane integrity and oxidation. The drying curves with varying temperatures could be well described with a mechanistic drying model while the shrinkage seemed to be non-ideal and related to the glassy state. The influence of temperature, RH and time on quality could be described with a highly significant regression model.

Conclusion:

Drying data combined with quality data have been collected indicating systematic drying effects on seeds. In addition, mechanistic and regression models were developed which will be used to predict optimal drying. This might lead to new best practices in industrial seed drying and hence more sustainable and long-lasting high-quality seeds.

Detection of meatball adulteration with pork meat using vibrational spectroscopy

Zaqlul Iqbal^{1,2}, Nils Kristian Afseth², Annelies Postelmans¹, Joni Kusnadi³, Wouter Saeys¹

¹Department of Biosystems, MeBioS division, KU Leuven, ²Faculty of Agricultural Technology, Universitas Brawijaya, ³Norwegian Institute for Food, Fisheries and Aquaculture Research

Aim:

Muslim-majority countries like Indonesia have to screen for adulteration of processed meat products such as meatballs with pork meat. Currently, the Halal authority relies on the analysis of DNA, protein, or fat with RT-PCR, LC-MS, or GC-FID. While these methods are very reliable, they are not suitable for rapid screening of large numbers of samples. Therefore, fast screening tools are needed to identify suspected samples which can be verified with these reference methods. Vibrational spectroscopy methods such as Raman and NIR spectroscopy are fast and non-destructive, and widely used in food analysis. As the acquired spectra provide a molecular fingerprint, the aim of this study was to identify their ability to detect pork adulteration in meatballs.

Method:

Two sample groups were prepared: 3 batches of adulterated samples (beef meatballs with 3, 5, 10, 50, and 100% pork adulteration) samples and 10 batches of pure beef meatballs. Each percentage had five meatballs prepared, totaling 125 samples. The samples were scanned intact and after cutting them in half (cross-section) in reflectance mode. The Kaiser RXN2 was used for Raman spectra (200 to 1809 cm^{-1}), and NIRsystems XDS Foss was used for NIR spectra (1100-2500nm). Partial least squares discriminant analysis (PLS-DA) models were trained to discriminate adulterated and non-adulterated meatballs. Around 2/3 of the samples were used for model calibration and the remaining 1/3 was used for model validation.

Results:

The acquired Raman spectra have dominant peaks at 1657 cm^{-1} , 1443 cm^{-1} , and 1299 cm^{-1} which were attributed to the saturated and unsaturated fat while absorption peaks for fat and lipids were observed in the NIR spectra at 1732 nm and 2312 nm. The accuracy of the PLS-DA models for discriminating adulterated from non-adulterated meatballs could reach up to 75% for Raman and NIR spectroscopy by using external validation. However, most samples with a low level of adulteration were misclassified as non-adulterated.

Conclusion:

Raman and NIR spectroscopy have potential as rapid screening tools for the detection of high adulteration rate. Future research is recommended to investigate the effect of the fat content of the beef and pork meat on the discrimination performance.

Radio Frequency Thermal Processing of Grounded Peanut Samples: Development of a Computational Model

MSC. STUDENT CANER TASCI¹, Msc. Student Elif Nida Cakir¹, Msc. Student Kubra Polat¹, Dr. Shuxiang Liu², Dr. Ferruh Erdogan¹, Dr. Samet Ozturk³

¹Department of Food Engineering, Ankara University, ²Institute of Food Processing and Safety, College of Food Science, Sichuan Agricultural University, ³Department of Food Engineering, Gümüşhane University

Aim:

There have been peanut butter related outbreaks and recalls by CDC and FDA within the last decade, mainly due to *Salmonella*, and this indicates an intermediate thermal process to be applied to peanuts for decontamination. Due to the grounding, particle size might be an important aspect in the view of thermal processing, and applying a novel approach has been another consideration due to the UN 2030 Sustainable Development Goals. Therefore, this study aimed to develop a mathematical model for predicting temperature distribution of grounded peanuts with respect to the particle size during radio frequency (RF) thermal decontamination process.

Method:

A free oscillating pilot-plant scale staggered through field RF system (10 kW, 27.12 MHz) was used at 100 mm electrode gap and 4000V potential until the centre temperature reached to 80°C. Peanut samples (160 g, placed in cylindrical polypropylene container; 75.60 mm in diameter and 67 mm in length) of four different sizes (2.00, 1.40, 1.00, and 0.71 mm) were used in the RF heating experiments. Temperature changes were measured by fiber optic sensors at 3 different locations (top, centre, and bottom) within the sample, and a mathematical model was developed using Comsol Multiphysics (V6.0) to determine the electromagnetic field distribution within the RF cavity and temperature distribution within the sample. Thermophysical properties of the peanut samples were calculated using the composition, and dielectric properties were experimentally measured as a function of temperature and frequency.

Result:

Mathematical model results compared well with the experimental data, and the model predictions revealed that porosity (due to the particle sizes) did not significantly affect the overall heating rate and temperature distribution of the samples. In fact, the porosity change was found to be not significant within the given particle size range. Using the experimentally validated model, then, industrial processing conditions with larger number of the samples were also demonstrated.

Conclusion:

The developed mathematical model provided a valuable tool for predicting heating rates and temperature distribution based on particle size during RF processing of various sized peanut samples, and industrial scale processing conditions were also determined.

Computational Modeling of Radio Frequency Thawing for Frozen Whole Liquid Eggs

MSc. Student Dilara Ulu¹, Ozan Karatas¹, Eda Coskun¹, Ozan Altin¹, Ferruh Erdogdu¹

¹Ankara University

Aim:

Due to the volume of laid eggs in poultry industry, excess cases might be stored. For this purpose, the whole shell eggs are frozen in the form of a whole liquid egg (WLE). Then, they should be thawed, and conventional thawing process takes longer times. With volumetric heating feature of radio frequency (RF) processing, it might offer a solution to this industrial problem, but designing such a process has its challenges with temperature uniformity as a function of non-uniform electromagnetic field distribution. Therefore, the objective of this study was to develop a computational model for radio frequency thawing of frozen WLEs and use this model to design an industrial scale process.

Method:

For this purpose, frozen WLE samples were obtained from a local producer. Following the measurement of dielectric properties and calculation of thermal-physical properties with respect to composition, an enthalpy approach based mathematical model was developed using Comsol multiphysics (V6.2). Experimental thawing studies were carried out in a staggered through electrode configuration 27.12 MHz 10 kW RF system. The samples were in size of 100×70×70 mm, frozen in boxes, and thawing was completed at 12 and 15 cm electrode gap, corresponded to 4500 and 5000 V potential, respectively. Temperature changes through RF thawing process were measured with fiber optic probes, and this data was used in model validation.

Results:

Enthalpy method based computational model results compared well with the experimental data obtained at two distinct points within the samples. The results indicated the significance of product depth on temperature evolution with noted non-uniformity. Hence, further simulation studies were carried out to determine the optimum sample size and process conditions for temperature uniformity. The results were also compared with the conventional (natural convection air-thawing) approach to demonstrate the significant effect of RF processing on thawing process time.

Conclusion:

The results of this study indicated the RF process as a potential novel approach for thawing of frozen WLEs. This approach offered an industrial scale application in the egg industry with the UN 2030 Sustainable Development Goals (SDGs) for the use of clean energy in food processing.

Microwave Processing of Ice Cream Mixes: System Design and Microbial Inactivation Evaluation

Msc Elif Nida Cakir¹, Ph.D. Student Eda Coskun¹, MSc. Student M.Talha Akbulut¹, Ph.D. Student Tugba Bulat², Dr. Tuncay Yilmaz³, Prof.Dr. S. Aykut Aytac², Prof.Dr. Ferruh Erdogdu¹

¹Department of Food Engineering, Ankara University, ²Department of Food Engineering, Hacettepe University, ³Department of Food Engineering, Manisa Celal Bayar University

Aim:

Ice cream is a high nutritional and economic value product, and it creates a favorable environment for microorganism development with its composition,. Ice cream-related outbreaks and recalls by CDC and FDA, mainly due to *Listeria monocytogenes*, have been observed.. Hence, it is of importance for ice cream mixtures to undergo an intermediate thermal process. In line with UN 2030 Sustainable Development Goals (SDGs) for the use of clean energy and reducing process carbon footprints, the objective of this study was to evaluate the microwave heating on microbial inactivation and develop a industrial scale microwave process for ice cream.

Method:

Ice cream mixture was obtained from a local manufacturer, and microwave heating experiments were carried out in a custom design 1 kW- 2450 MHz system. 550 g sample placed in a 9.6 cm diameter polypropylene cylinder was used in the experiments. For microbial inactivation studies following the inoculation of *L. monocytogenes* 1/2c ATCC 7644 strain culture, microwave processing was applied for 200, 400, 600, and 750 s, and color change of the ice cream samples were also measured. Based on the microbial inactivation data and corresponding temperature, a previously developed and experimentally validated model was used for designing a continuous flow industrial scale system.

Results:

Less than 1 log CFU/g decrease decrease was observed till 400 s while 2.70 log cycle reduction and a complete inactivation were obtained at 600 and 750 s, respectively. These process times corresponded to over 76 and 88 °C average sample temperature, and the validated model was used to determine the temperature evolution. No significant change was observed in color in terms of $L^*a^*b^*$ values. With respect to the required temperature for complete inactivation, continuous flow system design studies were presented for temperature uniformity and efficient processing.

Conclusion:

With the microwave processing, desired microbial inactivation was achieved while the color of the sample was preserved. Microwave processing was demonstrated with its potential as a sustainable approach, and industrial scale desing studies were demonstrated using the previously developed and validated computational model.

Microwave Processing of Butter Milk: Process Evaluation for Microbial Inactivation and Quality Attributes

Msc. Student Kubra Polat¹, PhD. Student Eda Coskun¹, BSc. Student Elif Beyza Kenger², Assoc. Prof. Dr. Eda Demirok Soncu¹, Prof. Dr. Sait Aykut Aytac², Prof. Dr. Behic Mert³, Prof. Dr. Ferruh Erdogdu¹
¹Department of Food Engineering, Ankara University, ²Department of Food Engineering, Hacettepe University, ³Department of Food Engineering, Middle East Technical University

Aim:

Butter milk is reported to be a potential source of *Salmonella* outbreaks in chocolate-derived products in recent years. In view of UN 2030 Sustainable Development Goals for clean energy and reducing process carbon footprints with novel approaches, the objective of this study was to develop a mathematical model to determine temperature evolution of butter milk during microwave heating and evaluate the process potential to inactivate *Salmonella* and other microorganisms.

Method:

Butter milk sample was obtained from a local plant. Microwave heating experiments were carried out in a custom design 1 kW- 2450 MHz system. A cylindrical polypropylene container was used to place 100 mL sample during processing, and temperature changes were recorded using fiber optic probes. Experimentally obtained temperature data was used for mathematical model validation, developed using Comsol Multiphysics v6.2. Dielectric and rheological properties were also measured for modeling approach. Following the validation, butter milk sample was inoculated with a culture containing *Salmonella* Enteritidis, ATCC 13076. Enumeration was carried out in the samples when the center temperature reached to 50 to 80 °C (these corresponded to 60 to 100 s) to determine coliform bacteria, total mesophilic aerobic bacteria, yeast-mold, total psychrophilic aerobic bacteria as well as *Salmonella* Enteritidis. Quality analysis included to assess emulsifying properties, color, phase separation, peroxide content, and detection of protein changes using gel electrophoresis.

Results:

The model results compared well with the experimental data. Microwave processing did not result in a significant change in emulsion stability, phase separation at 48 h, peroxide value and protein profile while significant reduction in b^* value was observed. Coliform number was decreased by 3-log while the reduction mesophiles and psychrophiles was 2-log and 1.5-log in yeast-mold number. Average temperature of 70 °C and 80 °C of the butter milk samples led to a complete inactivation of *Salmonella* Enteritidis indicating the microwave heating as an efficient application for thermal processing of butter milk.

Conclusion:

The results of this study are expected to contribute to use of novel processing approaches in the view of food safety, and the developed mathematical model might be used further to design industrial scale systems.