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Safety for future generations

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Plenary Session

Animal production and food quality

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ABSTRACT

Over the past century, animal production has undergone major changes, shifting from traditional, resource-based systems to intensified, technology-driven ones. These changes have primarily increased productivity but have also introduced environmental, economic, and societal challenges. Meanwhile, the concept of food quality has evolved and is now recognised as multidimensional. Food quality refers not only to nutritional value but also to safety, sensory characteristics, and consumer perception. Various factors, including production conditions, genetics, nutrition, animal health, and management practices, affect final product quality. Advances in genomics, precision livestock farming, and digital technologies offer new opportunities to improve efficiency and quality parameters. However, adoption of these technologies remains uneven. At the same time, growing consumer awareness of animal welfare, environmental sustainability, and product origin is reshaping expectations and influencing both market demand and regulatory frameworks. Recent global disruptions, including pandemics affecting both human and animal populations, have further exposed the vulnerability of highly globalised food systems and contributed to renewed interest in shorter supply chains, local production, and food system resilience. The relationship between animal production and food quality is therefore complex and multidimensional. Structural and technological changes in production systems affect productivity, consistency, diversity, and perceived quality of animal-derived foods. Addressing current and future challenges requires integrated approaches that balance efficiency and sustainability, while responding to evolving societal expectations.

Keywords: livestock systems, animal welfare, sustainability, nutritional value, food safety

Listening to Wetlands: Bioacoustics and AI for Biodiversity Monitoring in Kopački Rit Nature Park

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ABSTRACT

Acoustic monitoring is rapidly transforming biodiversity research by enabling continuous, non-invasive, and spatially extensive wildlife surveys. This presentation will explore the application of bioacoustics in biodiversity monitoring, using practical examples from the Tech4All (WatchOUT) project in Kopački Rit Nature Park, Croatia. Kopački Rit is one of Europe's most important floodplain wetland ecosystems, located at the confluence of the Danube and Drava rivers. Passive acoustic recorders were deployed across 95 sampling sites, generating more than 3.2 million one-minute recordings from September 2023 to the end of 2024. These data were analysed using pattern matching, artificial intelligence models, and ecological occupancy modelling. The acoustic surveys detected 96 species, including 88 birds, 4 mammals, and 4 amphibians. Among them were species of conservation concern, as well as ecologically important and flagship species including the white-tailed eagle, golden jackal, red deer, great spotted woodpecker, and European tree frog. The results demonstrate how bioacoustics can reveal not only species presence, but also spatial distribution, habitat associations, and daily activity patterns. Amphibians were strongly linked to wetland habitats and proximity to water, birds showed associations with mature forest structure and canopy height, while mammals were most frequently detected in ecotonal zones where forest, wetlands, and recovering vegetation intersect. Temporal analyses further showed distinct activity rhythms, from nocturnal calling in tree frogs to crepuscular activity in red deer and golden jackals. This study highlights the value of combining passive acoustic monitoring, AI-based species detection, remote sensing data, and ecological modelling for biodiversity assessment and conservation planning. The Tech4All (WatchOUT) project provides a scalable framework for long-term monitoring of dynamic wetland ecosystems and offers practical tools for adaptive management, habitat prioritisation, and future climate resilience strategies.

Keywords: occupancy modelling, species detection, acoustic datasets, habitat heterogeneity, ecological indicators

Acknowledgment: This research was supported by the Tech4All programme Nature Guardian, under project "Wetland and wildlife monitoring using technology and acoustics"

Youth and Education in Life Sciences: From Fragmentation to Growth

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ABSTRACT

This keynote explores how the New World Order, as articulated by Mark Carney, is reshaping economic development from centralised systems to flexible, networked cooperation. In this context, regions like Croatia face urgent challenges: an ageing agricultural workforce, rural depopulation, and the loss of added value through the export of raw commodities. At the same time, education systems lag behind rapid technological change, leaving young talent insufficiently prepared for modern labour markets.

The presentation argues that fragmentation between education, industry, research, and policy is the core barrier to growth. To address this, regions must build integrated, data-driven ecosystems that align talent with real-world demand and accelerate innovation and entrepreneurship. SMRT.BIO is introduced as a digital infrastructure that operationalises this shift, connecting individuals, employers, educational institutions, and policymakers into a single system.

Using the FAZOS Faculty of Agrobiotechnical Sciences Osijek as an example, the keynote demonstrates how institutions can evolve from knowledge providers to drivers of regional economic development. We focus on cross-regional youth collaboration between Europe, Africa, and Asia to bring diverse perspectives, solve global problems, and build long-term talent networks.

By linking skills development to entrepreneurship and economic outcomes, regions can move from fragmented potential to coordinated growth. The keynote concludes that competitiveness in life sciences will be defined by the ability to connect, collaborate, and execute at scale and speed, turning talent into measurable socio-economic impact

Keywords: digital transformation, regional economic, Data-driven ecosystems

Transforming bio-waste into innovative solutions for regenerative agroecosystems

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ABSTRACT

The growing accumulation of waste from diverse streams, coupled with climate change and increasing pressure on natural resources, presents a significant global challenge and an opportunity for innovation. Although substantial quantities of bio-waste materials remain underutilized or are disposed of in landfills, these materials represent valuable sources of organic matter and nutrients that can be recovered and reintegrated into agroecosystems. Innovative bio-waste valorization approaches are enabling the conversion of diverse waste matrices into high-value products (e.g., growing substrates, soil amendments, organic and mineral fertilizers) that support regenerative agriculture and are optimized to enhance the sustainability and productivity of the agri-food sector. These solutions contribute to improved soil health, enhanced microbial activity, increased water and nutrient use efficiency, while reducing reliance on synthetic inputs and finite natural resources, and protecting environmental resources from contamination. By closing nutrient and carbon loops, such innovations advance circular bio-economy principles and contribute to more resilient, climate-smart agroecosystems. Finally, these approaches align closely with European Union strategic priorities in waste valorization, soil health, climate adaptation, and resource efficiency, demonstrating how bio-waste conversion can reduce landfill dependency, improve waste management practices, and generate smart green innovations.

Keywords: climate change, agriculture, waste upcycling, food safety and security, resource management

Data is the New Harvest: Digital Agriculture Between Innovation, Ethics, and Croatian Reality

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ABSTRACT

Croatia's struggle to pursue digitalization of agriculture in spite of its potentially advantageous position is covered in this article. The goal is to critically assess the smart agriculture narrative as a 'solution' by looking at how data is made into a new type of value, by whom, and how to prevent the digital transition from deepening inequalities between farmers. According to recent European research, national data on farm structures and experiences from field trials, most European farms are using at least some specific technology. However, only a small share uses them in integrated systems. Adoption is heavily influenced by farm size, digital skills, and perceived profitability. In Croatia, where three quarters of agricultural holdings manage 5 ha or less, high upfront costs, a fragmented infrastructure, and low trust in data platforms create a digital divide threatening small-scale producers with marginalization. Research on technology acceptance indicates that farmers do not reject innovations through technophobia but through rational assessment of risk and benefits. The likelihood of adoption increases with articulated economic returns and clear data ownership models. Educational framework and public policies are often behind the tech cycles and do not help in trust building. If digital agriculture is technocentric and exclusionary, it will not become sustainable. To achieve a successful transition, it is not enough to build the infrastructure, but inclusive ecosystems based on ethical data governance, tailor-made capacity-building by way of policies, and the diverse character of Croatian agribusiness. In short, smart agriculture must balance between tradition and innovation, so that the farmer in the field is served by the data.

Keywords: digital agriculture, digitalization, data ethics, Croatian agribusiness, technology adoption

Session 1

Plant and Food Production

Awareness of dental erosion among Croatian certified wine assessors

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ABSTRACT

Professional wine tasters encounter a high risk of dental erosion as a consequence of frequent and intensive exposure of oral structures to acids in wine. Due to the professional hazard, they cannot evade, additional specific procedures in oral care are advised to prevent dental erosion as well as to alleviate its symptoms. The aim of this study was to determine the knowledge and awareness of certified Croatian wine assessors regarding dental erosion, its aetiology, and prevention. An online questionnaire was sent to 108 active certified Croatian wine assessors of which 83% responded. The information obtained was analysed using methods of descriptive statistics and tests commonly used in questionnaire-based studies. Although most wine assessors regularly use dental services, in one third of the cases dentists were not aware of their profession. The proportion of dentists familiar with their patients' occupation did not differ between the public and private healthcare sectors. While two thirds of participants reported having heard of the term “*dental erosion*”, their understanding and objective knowledge were limited. Furthermore, the level of knowledge was not associated with dentists' awareness of the participants' profession - minority reported dentists as their primary source of information about dental erosion. Most participants have been informed incidentally, mostly through various forms of media. The majority of wine assessors were not familiar with the appropriate oral care practices for preventing dental erosion and reported no specific hygiene before or after wine tasting. These behaviours did not vary with increasing professional experience. The knowledge and awareness of dental erosion among Croatian certified wine assessors was found to be insufficient as well as their oral care practices with regard to the increased risk for erosive tooth wear. The findings underscore the need for targeted education, both within routine dental practice and through the development of educational materials on dental erosion and its prevention for professional wine tasters.

Keywords: wine tasting, dental erosion, prevention

The potential of using sheep wool pellets in lettuce cultivation

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ABSTRACT

Although approximately 1,300 tons of wool are produced annually in Croatia, its disposal often presents a problem, and it is frequently discarded in nature, creating an environmental issue. However, in recent years, it has been processed into sheep wool pellets (SWP) and used in agriculture. A one-year fertilization experiment was conducted in greenhouse lettuce production, including various organic fertilizers commonly used in practice as comparative treatments. The aim of the study was to investigate the effect of SWP on soil chemical parameters, soil CO₂ emissions, as well as morphometric parameters, lettuce yield, and antioxidant capacity. Soil and plant material analyses were carried out, and the results showed no significant differences, except for plant height and yield, where BIO-N treatments had higher values compared to mature manure, while other treatments were comparable. The control treatment and manure showed higher antioxidant capacity in plant material according to the ORAC method. These results from a one-year study provide insight into the potential use of SWP in lettuce cultivation, showing comparable results to other organic fertilizers in many parameters; however, long-term effects on soil and the cultivation of different crops should be further investigated.

Keywords: sheep wool, lettuce, antioxidant capacity, fertilization, sheep wool pellets

The Effect of Water Salinity on Transpiration and Photochemistry in *Citrus unshiu* Marc. 'Ichumare' in Croatia

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ABSTRACT

The global climate is undergoing rapid changes, significantly affecting agricultural production worldwide. Rising sea levels and irrigation-related secondary salinization are increasing soil and water salinity, posing serious challenges for the cultivation of salt-sensitive crops such as citrus in Mediterranean regions, including Croatia. An experiment was conducted on the early mandarin cultivar 'Ichumare' during three growing seasons (2023–2025) at two locations in the Neretva Valley, Croatia (Vidrice and Bostanac), representing contrasting salinity conditions. The electrical conductivity (EC) of irrigation water was considerably higher at Vidrice compared with Bostanac, indicating stronger salinity stress at this location. Physiological parameters, including stomatal conductance (g_{sw}), quantum efficiency of photosystem II (PhiPSII), and relative electron transport rate (ETR) were measured using a LI-600 porometer/fluorometer in June, August, and October each year. In 2023, higher average stomatal conductance was recorded at Bostanac ($0.126 \text{ mol m}^{-2} \text{ s}^{-1}$) compared with Vidrice ($0.079 \text{ mol m}^{-2} \text{ s}^{-1}$). In contrast, higher values were observed at Vidrice in 2024 ($0.098 \text{ mol m}^{-2} \text{ s}^{-1}$) and 2025 ($0.134 \text{ mol m}^{-2} \text{ s}^{-1}$), while lower values were recorded at Bostanac (0.073 and $0.071 \text{ mol m}^{-2} \text{ s}^{-1}$, respectively). Quantum efficiency of photosystem II (PhiPSII) differed significantly between locations only in 2025, when higher values were observed at Vidrice. These findings suggest that salinity effects on mandarin physiology are strongly modulated by interannual environmental variability. The observed fluctuations in stomatal conductance, coupled with relatively stable photochemical parameters, may indicate partial physiological acclimation of 'Ichumare' to saline conditions. However, the inconsistent response across years highlights the complexity of salinity stress under field conditions.

Keywords: climate change, salinity, mandarin, stomatal conductance, fluorescence

Project: “Water, soil and cultivar - fundamental elements of sustainable agriculture in climate change (VoTiKult)”

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ABSTRACT

In modern agricultural production, one of the biggest challenges is climate change, which is increasingly affecting agricultural production. Achieving high yields and quality in order to maintain competitiveness is an increasing challenge for most producers while production costs are rising.

The paper presents the results of a four-year research into the quality of three different fractions of soybean seeds (Ika, Sonja and Korana) and wheat (Indira, Barba and Kraljica). The chemical composition of soybean and wheat seed fractions was analyzed and the qualitative properties of soybean and wheat seed fractions were determined. Photosynthetic activity was investigated in the vegetative phase of development under optimal water regime conditions and under water stress conditions. Photosynthetic activity was measured on wheat leaves in the vegetative growth phase at optimal soil water content and under water stress, measured by various soil moisture methods. Water stress was monitored by analyzing proline and MDA concentrations as a plant response to stress.

The aim of the research was to determine the relationships between seed fractions and the influence of water regime on the properties of wheat and soybean. Differences in the chemical composition of seed fractions were visible in soybean and wheat. In soybean seeds, the average protein content was 40.09% and the average oil content was 21.68%, while the hectoliter was 71.69 g and the mass of 1000 grains was 159.52 g. In wheat seeds, the average protein content was 12.84% and the average starch content was 76.89%, while the hectoliter was 83.42 g and the mass of 1000 grains was 21.68 g. The differences in the photosynthetic activity of soybean and wheat plants after stress caused by lack of soil moisture were visible and statistically significant.

If smaller seed fractions do not differ from larger seed fractions in many properties, seed yield for sowing could be increased. In emergency situations of seed shortage (drought, natural disasters and catastrophes), the shortage can be alleviated by sowing smaller seed fractions. The results obtained by initiating drought and determining the intensity of drought in wheat and soybean can be linked to climate change.

Keywords: stress, drought, wheat, soybean, seed fractions

Long-term evolution of drought severity in Romania with a focus on grapevine development

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ABSTRACT

Drought assessment in Romania since 1961 is documented, but studies spanning on longer periods are few, and use either modelled or sparse observational data. Here we present a 123-year analysis of drought, water balance and aridity over Romania using data from 156 weather stations from the RoCliHom dataset, covering the period 1901–2023. Drought is analysed with the Standardized Precipitation Index (SPI) and Standardized Precipitation Evapotranspiration Index (SPEI) indices, while the aridity is investigated with the De Martonne Aridity Index. The non-parametric Mann-Kendall test is used for trend analysis, allowing a fair comparison with the most studies on Romanian aridity and drought. Trend magnitude is computed with Sen's slope estimator (Kendall-Theil robust line).

Our results show that the annual air temperature is increasing at all stations, as monthly temperature is rising in winter, spring and throughout the entire summer since 1901. The monthly temperature range is decreasing, suggesting that the thermal variability has diminished. The annual precipitation had slightly increased at some stations in southwestern, northeastern and southeastern Romania, but most of the stations present no significant trend. Potential evapotranspiration presents increasing trends in all seasons except autumn. SPEI showed an intensification of drought during the growing season (April-September) for the last 70, 60 and 50 years, yet longer periods show no trend. The De Martonne Aridity Index has significant trends at 9% of the stations, indicating a stable situation over the 123-year period. Water balance is declining in July and August, at most stations, which had certainly affected the ripening of grapevine and other autumn crops.

This work was supported by a grant of the Ministry of Research, Innovation and Digitization, CNCS-UEFISCDI, within PNCDI IV; project name: «Changes in climate suitability of Romanian vineyards» (CARVE); project number PN-IV-P1-PCE-2023-1875.

Keywords: vineyards, climate impact, weather station data, teleconnections, South-East Europe

Specific technologies in wine production and their labeling

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ABSTRACT

Specific winemaking technologies enable a shift from standardised wine quality and may serve as a valuable tool for market differentiation and promotion. The terms used to describe specific production technologies are optional labelling elements and may be used if the relevant production practices have been duly declared and are officially verified. Depending on the applied technology, such wines may have distinctive sensory profiles and quality attributes that set them apart from standard-produced wines. Within the European Union and the Republic of Croatia, the labelling of specific production technologies is allowed for wines with a Designation of Origin (PDO, PGI), and their recognisability is assessed within the official pre-market certification procedure. All wines with a PDO in Croatia undergo sensory evaluation using the 100-point method. In addition to numerical scoring, assessors are required to evaluate the recognisability of the specific production technology by descriptive answer, in accordance with national regulations and internal standards. The aim of this study was to identify which specific wine production technologies Croatian producers most frequently declare for labelling purposes and to analyse their frequency regarding some quality factors. The factors included in the data analyses were geographical origin (zone, PDO), grape variety, and wine color. The tested period was from 2023 to 2025, using data derived from internal databases and proprietary software. It was observed that the most frequently used terms referring to specific production technologies were as follows: oak ageing, (with various synonymous terms), extended maceration, lees ageing, and the labeling of unfiltered wines. Technologies involving oak ageing were significantly more prevalent in the production of red wines and viticultural zones C II and C III, whereas lees ageing was more commonly associated with white wines and production in all zones, especially in the C II zone. Terms related to the orange wine production and/or the use of amphora were less frequent. A considerable share of wines produced using specific technologies is linked to the traditional term ‘Vrhunsko vino KZP’, contributing to quality complexity and diversity within the wine market.

Keywords: wine labelling, PDO, specific technologies, wine market

Effect of fertilization and weather conditions on the economical production of sugar beet root mass and health significance

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ABSTRACT

One of the largest issues facing agricultural productivity is climate change. High temperatures and low amounts of precipitation cause large yield losses but also lead to certain plant diseases. The two macronutrients that are most necessary to sustain high sugar beetroot yields are nitrogen and potassium. Determining which NPK nutrient dosages produce the best outcomes is crucial since an excessive amount of nitrogen in the plant hinders the extraction of sugar. The Severina sugar beet variety was tested for three years on the experimental plots of the PSS Institute Tamiš. The impact of varying NPK fertilizer concentrations on root weight was investigated. Severina variety received supplemental fertilization with NPK nutrients (N₀P₀K₀, N₁₀₀P₀K₀, N₀P₁₀₀K₀, N₀P₀K₁₀₀, N₅₀P₅₀K₅₀, N₁₀₀P₅₀K₅₀, N₁₀₀P₁₀₀K₁₀₀, N₁₃₀P₅₀K₅₀, N₁₃₀P₁₀₀K₁₀₀, and N₁₃₀P₁₃₀K₁₃₀) to boost root output. The first year under examination had the largest root weight, whereas the third year had the smallest. The average weight of the roots was greatly impacted by the change in increasing NPK nutrition levels and year. On average, the variant N₁₃₀P₁₃₀K₁₃₀ had the biggest increase in root weight. The variant N₁₃₀P₁₀₀K₁₀₀ had the largest root weight in its first year. Climate change caused by warming is the main cause of global changes that significantly affect agricultural productivity. One of the main indicators of climate change is an increase in average temperatures. High temperatures lower product yield and quality when combined with prolonged periods of little precipitation or drought. Sugar beet production is especially affected by climate change.

Globally, the amount of sugar and other sweets consumed per person is rising. An average person should eat between 40-80 g of fat, 300-400 g of carbohydrates, and 120 g of protein each day. Because excessive consumption can result in long-term health problems like diabetes, obesity, and cardiovascular illnesses, this tendency is concerning. Current research actively discusses the health implications of refined sugar and sweeteners used as sugar substitutes. Dried whole beet, as an alternative to refined sugar, contributes to fiber, protein, and minerals in addition to its high sugar content and has great importance for health.

Keywords: sugar beet, root mass, NPK, climate change

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Introduction of poppy (*Papaver somniferum* L.) into *in vitro* culture

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ABSTRACT

Poppy (*Papaver somniferum* L.) is an important species in agriculture, pharmaceuticals, and research, but its cultivation is declining, highlighting the need to preserve genetic diversity. The population used in this study was collected at the Gradište site and recorded in the Croatian Plant Genetic Resources Database (accession IND00081). The aim of this study was to establish an *in vitro* culture from this population. Aseptic culture was established from seeds using NaOCl and initiation on hormone-free MS medium. After 30 days, a high percentage of viable plants (91%) was recorded. Plants were transferred to MS medium without hormones (T0) and to treatments with BAP + IBA + GA₃ (T1) and mT (T2), with the addition of iron in the chelated form FeEDDHA in all treatments. After 30 days, chlorosis was observed (20 – 90%). The lowest number of dead leaves was in T2 (3.22), while the highest shoot length was in T0 (4.1 cm). Embryogenic structures were observed in T1 and T2. After subculture, T0 showed only shoot elongation, T1 induced callus formation and moderate multiplication, while T2 showed strong shoot induction (85% of explants). The obtained results indicate a significant influence of the applied plant growth regulators on morphogenesis and the regenerative potential of poppy, with the meta-topolin treatment emerging as particularly promising for further protocol optimization. Future research will focus on the precise optimization of growth regulator concentrations in order to develop an efficient and reproducible micropropagation model for this species.

Keywords: controlled conditions, *Papaver somniferum*, micropropagation, plant growth regulators

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Etiology of pear trunk necrosis in Serbia

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ABSTRACT

Pears (*Pyrus communis*) are a nutritious fruit rich in vitamins C, K, and B6, as well as essential minerals such as potassium, magnesium, calcium, and dietary fiber. With a production of more than 48.000 tons in 2023, pears are one of the most important fruit crops in Serbia. In 2020, pear growers in the locality of Smederevo reported symptoms of trunk necrosis, including reduced growth and internal tissue discoloration. Eleven single-spore isolates with a uniform appearance were obtained, all forming dark gray colonies on PDA within seven days when incubated at 22 °C under a 12-hour photoperiod. The isolates exhibited rapid growth, with an average rate of 22.8 mm per day, and produced black microsclerotia (75 – 150 × 75 – 125 µm). The morphological features resembled those of *Macrophomina phaseolina*, which was further confirmed by ITS rDNA sequencing. A BLAST analysis of the ITS rDNA sequence of isolate 378-20 (Acc. No. PV252061) revealed 100 % identity with 34 isolates of *Macrophomina phaseolina* from different host plants worldwide. Pathogenicity was confirmed using the selected isolate (378-20) on healthy pear branches, as necrotic lesions appeared 15 days post-wound inoculation, whereas control plants remained symptomless. Successful reisolation was performed from all inoculated branches, fulfilling Koch's postulates. This study presents the first report of *Macrophomina phaseolina* causing pear branch necrosis in Serbia, which is very important for effective disease management strategies, particularly in young orchards. This paper is the result of projects 451-03-34/2026-03/200116 funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia.

Keywords: pear, *Macrophomina*, identification, trunk necrosis, pathogenicity

Detection of Viruses in Fig (*Ficus carica* L.) in Croatia Using High-Throughput Sequencing

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ABSTRACT

Fig (*Ficus carica* L.) is an economically and culturally important fruit crop in Mediterranean regions, where perennial plants are frequently exposed to complex pathogen pressure. The aim of this study was to detect viruses infecting fig trees in the coastal part of Croatia using high-throughput sequencing (HTS). Six fig trees were sampled from two locations: Kaštela (4) and the Pelješac Peninsula (2), irrespective of symptom expression. Total RNA was extracted individually using a modified RNeasy Plant Mini protocol optimized for woody plants and subsequently pooled according to RNA concentration. The pooled RNA was DNase-treated, and its concentration, purity, and RNA integrity number (RIN) were determined prior to sequencing on the Illumina NovaSeq X Plus platform using paired-end reads (2 × 150 bp). Raw sequencing reads were quality-filtered and trimmed before de novo assembly using Trinity. Assembled contigs were analysed by BLAST against the NCBI database, and virus-related reads were mapped to reference genomes using Geneious Prime to confirm sequence identity and genome coverage. Bioinformatic analysis of the Illumina dataset revealed the presence of nine plant viruses: fig badnavirus 1, fig leaf mottle-associated viruses 1 and 2, fig mild mottle-associated virus, fig mosaic virus, fig virus B and fig virus C, an unclassified “fig umbra-like virus” and a ficus tepovirus A/cherry virus T. All identified viruses were confirmed by virus-specific PCR assays, demonstrating complex mixed infections in some of the analysed fig material. The detection of multiple economically important viruses in a limited number of plants, including asymptomatic individuals, highlights the need for regular monitoring and reliable diagnostic approaches in perennial fruit production systems.

Keywords: fig, virus detection, high-throughput sequencing

Greening the City as an Urban Ecosystem: An Integrated Model for Planning and Managing Urban Green Spaces

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ABSTRACT

Contemporary cities, including Zagreb, face increasing urbanization pressures, climate change impacts, and a decline in available green spaces, while traditional sectoral planning approaches often fail to ensure effective management of urban ecosystems and the optimal use of ecosystem services. This paper aims to develop and present an integrated model for planning and managing urban green spaces, based on the concept of the city as a dynamic and interconnected ecosystem. The proposed model combines spatial planning, ecological indicators, climate parameters, and participatory governance mechanisms. It includes a comprehensive analysis of the existing situation, identification of priority areas for greening, the establishment and enhancement of green corridors, and the development of a long-term management and monitoring framework. The results show that this approach contributes to increased climate resilience, mitigation of urban heat island effects, improved air quality, and enhanced quality of life. Furthermore, the model supports data-driven decision-making and fosters interdisciplinary cooperation and stakeholder engagement. The findings indicate that the integrated model provides a strong basis for the development of a functionally connected, climate-resilient, and socially inclusive urban ecosystem, while offering a transferable and scalable framework for sustainable urban planning in comparable urban environments.

Keywords: urban greening, urban ecosystem, green infrastructure, climate resilience, participatory planning

Photosynthetic response to foliar zinc nutrition in *Vitis vinifera* L. cv. Syrah

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ABSTRACT

Agronomic biofortification is a recognized strategy for mitigating micronutrient deficiencies, particularly zinc. Zinc is indispensable for proper human development and the maintenance of health. Within this framework, an agronomic Zn-enrichment workflow was implemented in irrigated *Vitis vinifera* cv. Syrah to assess the mobilization of photoassimilates toward the grapes and to determine its impact on total soluble solids accumulation. Throughout the production cycle, four foliar applications were carried out using ZnSO₄ or ZnO at a concentration of 1350 g·ha⁻¹. At a physiological level, gas-exchange photosynthetic parameters and the normalized vegetation index showed no evidence of Zn toxicity. Moreover, it is worth noting that vines treated with ZnSO₄ even exhibited a slight, although non-significant, increase in net photosynthesis (P_n), suggesting a potential benefit. At harvest, vines treated with ZnSO₄ and ZnO showed an increase in Zn content, with the latter displaying a 1.2-fold increase relative to the control. Furthermore, both ZnSO₄ and ZnO foliar applications significantly enhanced the total soluble sugar content of the grapes at harvest. In conclusion, foliar application of ZnO and ZnSO₄ proved effective, offering potential benefits for winemaking while showing no clear signs of adverse effects.

Keywords: biofortification, photosynthetic parameters, *Vitis vinifera* L., zinc.

Impact of Foliar Protein Hydrolysates Application on the Phenolic Profile of Olive Seedlings

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ABSTRACT

Protein hydrolysates (PHs), obtained by enzymatic degradation of plant proteins from agro-industrial plant waste, are biostimulants that can be used in sustainable olive cultivation. Therefore, this study investigated the effects of PHs foliar application on the leaf phenolic concentration of ‘Buža’ and ‘Leccino’, two economically important olive cultivars in the Istrian region. Over eight months in the greenhouse, one-year-old olive seedlings received standard nursery irrigation and were supplemented periodically with foliar treatments of water (control) or three different PHs derived from pumpkin seed cake: H1 (Alcalase[®] Pure), H2 (Alcalase[®] Pure + Flavourzyme[®]), and H3 (Alcalase[®] Pure + Protana[™] Prime). At the end of the experiment, the results showed that the H1 treatment significantly increased the concentrations of oleuropein and oleuroside compared with the control. Other reactive phenolic secoiridoids, often associated with plant defense-related phenolic metabolism, such as oleacein and oleocanthal, were markedly higher in the control treatment than in all applied PH treatments. Verbascoside showed a cultivar x treatment-dependent response, with the highest values in ‘Buža’ under H1 and the lowest in ‘Leccino’ under H3 treatment. This study indicates the potential of PHs' foliar application as an environmentally friendly practice and provides new insights into treatment vs. cultivar-specific shifts in olive secondary metabolism.

Keywords: pumpkin seed cake, enzymatic degradation, biostimulants, ‘Buža’, ‘Leccino’

The effects of organic growing technology on the productivity of spelt

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ABSTRACT

The main objectives were to examine the effects of technology based on organic principles on the morphological and productive characteristics of spelt (*Triticum aestivum* L. ssp. *spelta*), cultivar Nirvana. This paper presents results from the investigated period (2020/21–2021/22) on chernozem luvic soil at the "Radmilovac" Experimental Field, Faculty of Agriculture. The trial was part of a four-crop rotation (maize, winter wheat, spring barley + red clover, red clover second year). In this rotation, red clover was used as an additional source of nitrogen. Conventional tillage with a ploughshare was performed at a depth of 25 cm in mid-October in both years, followed by seedbed preparation with a disc harrow and harrow immediately afterwards.

The experiment examined: organic fertiliser applied immediately before primary tillage at a rate of 30 t ha⁻¹; soil conditioners incorporated into the soil in autumn along with pre-sowing preparation (500, 150, and 300 kg ha⁻¹); fertilisation with organic and microbiological products during top-dressing; and fertilisation with soil conditioners and microbiological products during top-dressing.

The organic fertiliser under the trade name "Royal Biohumus ofert" is characterised by a high pH of 8, and average contents of N 2.1%, P₂O₅ 3.6%, and K₂O 2.2%. The soil conditioner Zeo pullus is characterised by average contents of N 6%, P₂O₅ 6%, K₂O 6%, CaO 8%, MgO 3%, Fe, Cu, and Zn; Zeo mag (MgO 17%, Zn 3%, and Mn 3%) and Zeo fos (P₂O₅ 4–5%, MgO 17%, and CaO 40%). "Slavol" is a natural microbial fertiliser containing two sets of nitrogen-fixing bacteria and phosphorus/mineralisers, as well as some biostimulators.

The combination of organic and microbiological fertiliser had a better effect on the morphological characteristics and grain yield of spelt in 2020/21. However, in the drier second year of investigation, the advantage of soil conditioners was established, especially in grain yield. The average grain yield of spelt was 4.44 t ha⁻¹, with the highest yield recorded in treatments with organic fertiliser and Slavol in the first year of investigation (5.12 t ha⁻¹).

Keywords: genotypes, alternative small grains, organic farming, yield

From agro-waste to value: onion peels as a sustainable source of bioactive compounds

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ABSTRACT

Onion (*Allium cepa* L.) is one of the most widely cultivated vegetable crops in Europe and worldwide. Its increasing production generates substantial amounts of solid waste, particularly onion peels. These by-products are recognized as a rich source of bioactive compounds with significant antioxidant and antimicrobial potential. The objective of this study was to evaluate the chemical composition, antioxidant capacity, and antimicrobial activity of extracts obtained from red and yellow onion peels as agro-industrial by-products. Extracts were prepared from dried, pulverized peels of different particle sizes (>500 µm and <500 µm) using 50 % ethanol assisted by ultrasound (UAE) at a solid-to-solvent ratio of 10 g/200 mL. The extracts were analyzed for total phenolic content (TPC), chemical composition by HPLC, and antioxidant activity using assays that differ in their underlying reaction mechanisms (FRAP, DPPH, and ORAC). Antimicrobial activity was evaluated against selected Gram-positive and Gram-negative foodborne and spoilage bacteria. The results showed a high total phenolic content ranging from 1533.50 to 2188.00 mg GAE L⁻¹, with the highest value observed in the yellow onion sample with a larger particle size. HPLC analysis showed a predominance of flavonoids, particularly quercetin and its derivatives, as well as protocatechuic acid. Antioxidant assays confirmed strong activity of the extracts, with the highest FRAP (reflecting the reduction of Fe³⁺ to Fe²⁺) and DPPH (radical scavenging activity) inhibition values of 75.85 mM TE (Trolox equivalents) and 48.18%, respectively, in yellow onion peel extracts of smaller particle size (<500 µm). ORAC (ability of antioxidants to inhibit peroxy radical-induced oxidation) values ranged from 388.07 to 1380.19 mM TE, indicating considerable variability among samples and no clear correlation with particle size. Furthermore, the extracts demonstrated strong antimicrobial activity, especially against Gram-positive bacteria, with the lowest inhibitory and bactericidal concentration of 0.16 mg mL⁻¹ against *Bacillus cereus*. Onion peel by-products represent a valuable source of phenolic compounds with pronounced antioxidant and antimicrobial properties. These findings highlight their potential application as natural alternatives to synthetic additives in the food industry and beyond.

Keywords: onion by-products, phenolic compounds, antioxidant activity, antimicrobial activity, HPLC

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Biocontrol Potential of Bacteriophages Infecting *Erwinia amylovora* for Sustainable Fire Blight Control

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ABSTRACT

Fire blight, caused by *Erwinia amylovora*, represents a major challenge in pome fruit production, particularly under environmental conditions favourable for disease development. Biological control strategies offer a promising alternative to conventional bactericides with limited efficacy. During the period 2010–2024, a total of 34 bacteriophages infecting *E. amylovora* were isolated from enriched samples of soil, water, and symptomatic as well as asymptomatic plant tissue. Among them, four polyvalent phages were selected for detailed characterization, including morphological, physiological, and genomic analyses, as well as evaluation of their potential application in controlling fire blight *in vitro*. The selected phages produced clear or moderately clear plaques on bacterial lawns of different *E. amylovora* strains after 24 h of incubation. They were able to lyse 104 *E. amylovora* strains isolated from seven host plants in Serbia, as well as 19 *Pantoea agglomerans* strains originating from the rhizosphere and phyllosphere of apple and pear trees. Transmission electron microscopy revealed that all phages belong to the class *Caudoviricetes*, with myovirus-like morphology. Whole-genome sequencing showed genome sizes of 53 kb (three phage strains) and 162 kb (one phage). All genomes contained endolysin genes, with frequent presence of holin and spanin genes. No genes associated with lysogeny were detected. All phages effectively reduced *E. amylovora* populations in liquid culture, with significant differences observed in growth kinetics between untreated control and phage-treated cells. However, phage cocktails containing two or more phage strains were more effective in inhibiting bacterial growth than single-strain applications. The lytic activity and broad host range of these phage strains make them promising biocontrol agents against *E. amylovora*, while their application as phage cocktails may further improve treatment efficacy.

Keywords: biocontrol, bacteriophages, fire blight, host range, efficacy

Acknowledgement: This research was supported by the Science Fund of the Republic of Serbia, GRANT No. 7421, Innovative solutions in phage-mediated biocontrol of fire blight – InnovaPhage, and by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, Contract Nos. 451-03-33/2026-03/200010 and 451-03-33/2026-03/200116.

Potential of *Bacillus amyloliquefaciens* as a biostimulant for onion (*Allium cepa* L.) production

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ABSTRACT

Onion (*Allium cepa* L.) is one of the most economically important vegetable crops worldwide, recognized for its nutritional value and storage potential. The quality and storage longevity of onion bulbs are influenced by their biochemical composition and physiological condition. The aim of this study was to evaluate the effect of *Bacillus amyloliquefaciens* on the chemical composition and potential storage quality of onion bulbs. *B. amyloliquefaciens* was tested as an onion bulb pre-planting treatment and subsequent foliar treatments. The experiment was conducted on the onion cultivar Stuttgarter Riesen in two variants: treatment with *B. amyloliquefaciens* (ABO2) and the untreated control. Each variant consisted of three replicates of 200 plants, resulting in 600 onion plants per variant. The bacterium was initially grown in nutrient broth for 24 h, then transferred to a yeast based nutrient medium and cultivated for 70 h. Before planting, bulbs were immersed in the cultivation broth, and three foliar treatments were applied monthly during the growing season. The results showed that the application of *B. amyloliquefaciens* increased the onion bulb yield by 13.5% compared to the untreated control. The analyzed qualitative parameters indicated a slight improvement in the quality of treated bulbs: the total dry matter increased by 3.5% and the soluble solids content increased by 1.5%. No significant difference was observed in the ash content between the treated variant and the control, suggesting that the application of the bioagent did not negatively affect mineral composition of the bulbs. The results demonstrate that *B. amyloliquefaciens* (ABO2) positively influences both the yield and quality parameters of onion bulbs, particularly those related to postharvest stability and storage potential, indicating a biostimulatory effect. The moderate increase in dry matter and soluble solids content, combined with stable mineral composition, suggest that the treated bulbs are well suited for long-term storage. These findings emphasize the dual benefit of this bacterium in onion production and confirm its potential as a promising and sustainable biostimulant for agricultural application.

Keywords: *Bacillus amyloliquefaciens*, *Allium cepa*, biostimulation

Acknowledgment: The research funds were provided by the Ministry of Science, Technological Development and Innovation under Contract No. 451-03-33/2026-03/200117 dated 05 February 2026.

Assessment of the susceptibility of conventional and biofortified wheat grains to the rice weevil *Sitophilus oryzae* L. (Coleoptera: Curculionidae)

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ABSTRACT

The rice weevil (*Sitophilus oryzae* L.) is one of the most significant and destructive pests of stored grains, causing significant losses in grain quality as the primary pest. The study aimed to evaluate the susceptibility of the six wheat varieties (Dekan, Soissons, Ilirija, Srpanjka, Antonius, and Felix), grown conventionally and biofortified (enriched with selenium and zinc), to obtain more information on wheat variety susceptibility to rice weevil infestation. The number of F1 progeny and the length of the median developmental time of adult *S. oryzae* were monitored, and the resulting grain damage was assessed. The highest parental mortality of *S. oryzae* was recorded in varieties with the highest grain hardness. In conventional varieties, the highest mortality was recorded in the cultivar Dekan (26.3%), followed by Antonius (16.3%) and Ilirija (11.3%), while in biofortified varieties, the highest mortality was determined in Dekan (21.3%), Felix (18.3%), and Ilirija (11.3%). Consequently, a lower number of progeny was developed in these same varieties. The total grain weight loss in conventional varieties ranged from 0.6% (Dekan) to 3.72% (Felix), whereas in biofortified varieties, the losses were lower, ranging from 0.6% (Felix) to 1.8% (Dekan). By assessing damage in a random sample of 30 grains, all biofortified varieties (except Antonius) showed lower grain weight loss and a lower percentage of damaged grains compared to the same conventional varieties, with statistically significantly less damage observed in the Dekan variety. Based on the length of the median developmental time and the susceptibility index (DIS), wheat varieties were classified according to their susceptibility to rice weevil development. In conventional varieties, Dekan and Ilirija were found to be highly resistant, Antonius was resistant, Srpanjka and Soissons were moderately resistant, while Felix was moderately susceptible to the attack. The biofortified varieties showed a higher resistance category and stood out as highly resistant (Dekan) or resistant (Srpanjka, Felix, Antonius, and Soissons). These findings can help select wheat varieties suitable for long-term storage to minimize potential losses from major storage pest.

Keywords: wheat, stored-product pest, biofortification, susceptibility index, grain damage

Morphological and biochemical characterization of the traditional collard “Brsečka broskva”

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ABSTRACT

In the Brseč area, a traditional population of collard green (*Brassica oleracea* var. *acephala*) known as “Brsečka broskva” is cultivated on small plots and maintained through multigenerational on-farm conservation of local seed, with open pollination contributing to variability within the population. The aim of this study was to characterize the morphological and biochemical traits of “Brsečka broskva” using qualitative and quantitative descriptors as a basis for its conservation and valorization. The morphological description was carried out according to IBPGR descriptors for the genus *Brassica* and covered 22 traits, including morphotype uniformity, plant growth habit, leaf shape, incision and blistering, leaf colour, and the colour of petiole and stem, as well as the ability to form a head. Quantitatively, plant height and diameter, number of leaves, leaf blade length and width, and stem length and width were determined. A typical “Brsečka broskva” plant is semi-spreading, does not form a head, has obovate, moderately incised and moderately blistered green leaves, and a light green petiole and stem. Biochemical analysis of leaves included total polyphenols, glucosinolates and antioxidant capacity, with the obtained values confirming a high antioxidant potential of this population. The results indicate that “Brsečka broskva” represents a valuable local plant genetic resource with distinctive morphological traits and a favourable biochemical profile, providing a scientific basis for its conservation, future breeding research and the possibility of protection as a designation of origin.

Keywords: *Brassica oleracea*, plant genetic resources, morphotype, IBPGR descriptors

High-resolution Chloroplast SNV Analysis of diverse Grapevine (*Vitis vinifera* L.) Cultivars

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ABSTRACT

Grapevine (*Vitis vinifera* L.) is an important fruit crop with high genetic diversity, domesticated 6000–8000 years ago from wild *V. vinifera* ssp. *sylvestris*. This process led to key traits such as hermaphroditism and increased berry size. Owing to its complex diversity, molecular markers such as single nucleotide variants (SNVs), combined with next-generation sequencing (NGS), enable detailed analyses of genetic relationships. Chloroplast DNA (cpDNA) provides valuable insight into maternal lineages and complements nuclear genomic data. In this study, chloroplast SNVs were analyzed in 409 cultivated *V. vinifera* accessions using whole-genome shotgun sequencing (Ion Torrent technology) to investigate genetic diversity and phylogenetic relationships. The analyzed accessions originated from nine countries across Southeastern and Central Europe, as well as from a heterogeneous set maintained by INRAE. Sequencing generated 422.5 million reads, with an average chloroplast genome coverage of 87.6× and approximately 8.5% of reads mapping to cpDNA. A total of 93 SNVs were identified, most of which were shared across datasets, alongside several region-specific variants. Three haplotypes (ATA, ATT, GTA) were detected, with ATT being the most frequent (46.7%). Haplotype distribution differed significantly by geographic origin ($p = 0.000498$), with ATT predominating in the eastern Mediterranean, ATA in Western Europe, and GTA mainly within a heterogeneous group of varieties from a INRAE collection. Phylogenetic and network analyses revealed three major clusters corresponding to the identified haplotypes. While many chloroplast sequences were unique to single accessions, others exhibited clear regional clustering, reflecting human-mediated dispersal of grapevine cultivars through historical viticultural practices. These results demonstrate that cpSNV markers effectively reveal maternal lineage diversity and geographic structure in cultivated grapevine and represent a powerful tool for population-level studies in perennial crops.

Keywords: *Vitis vinifera*, cpDNA, SNVs, NGS, genetic diversity

The assessment of stability of the kernel number per row in maize inbred lines using nonparametric methods

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ABSTRACT

In this study, the stability of kernel number per row was assessed in 12 maize inbred lines with three different types of cytoplasm: cytoplasmic male sterility type C (cms-C), type S (cms-S), and fertile cytoplasm. Field trials were conducted during 2020 and 2021 at two locations in Zemun Polje (Selection Field and School Farm) using a randomized block design with three replications for each cytoplasm type. Analysis of mean values showed that lines with C-type cytoplasm had an average of 20.67 kernels per row, those with S-type cytoplasm had 19.85, while fertile lines achieved an average of 20.08 kernels per row. The range of average ranks for lines with C cytoplasm varied from 1.00 to 9.60, and values of the average rank difference across environments ($Si^{(1)}$) ranged from 0.00 to 4.50, while the rank variance ($Si^{(2)}$) varied between 0.00 and 14.33. The significance test for the average rank difference ($Zi^{(1)} = 17.9$) and rank variance ($Zi^{(2)} = 10.6$) was below the critical value (21.03), confirming that there were no significant differences in phenotypic stability among the genotypes and that no additional individual testing was necessary. Among lines with S-type cytoplasm, the highest average number of kernels per row was achieved by line L₁₀ (26.03), which also exhibited the highest stability ($Si^{(1)} = 0.67$), while the least stable line was L₁₁ ($Si^{(1)} = 5.83$). In fertile lines, the average number of kernels per row ranged from 15.70 (L₅) to 22.73 (L₂). The most stable line was L₅ ($Si^{(1)} = 0.50$), and the least stable was L₁₁ ($Si^{(1)} = 6.25$). The obtained results indicate the presence of variability in both the expression and stability of the tested genotypes, which can serve as a basis for further breeding work and the selection of stable lines with different cytoplasm types suitable for use in hybrid programs.

Keywords: maize, inbred lines, number of kernels per row, stability, cytoplasm

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Corn snack products enriched with wholemeal spelt semolina

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ABSTRACT

Extruded snack products are among the most popular snacks consumed worldwide. These products are rich in carbohydrates, fats, and salt. The main raw material for their production is corn grits. To improve nutritional value and physical and chemical properties, flours from different grains can be added to corn grits. Wholemeal spelt semolina is particularly notable for its superior nutritional profile, characterised by high concentrations of essential proteins, dietary fibre, and bioavailable minerals, which significantly enhance the functional value and sensory appeal of extruded corn snacks.

The aim of this study was to investigate the influence of wholemeal spelt semolina addition (in proportions of 4%, 8%, and 12% dry matter) on the properties of corn snack extrudates. The mixtures prior to extrusion were prepared at 15% moisture and extruded in a laboratory single-screw extruder. Samples were dried at room temperature and the following parameters were determined: expansion ratio, bulk density, colour, hardness, and fracturability of extrudates.

The addition of wholemeal spelt semolina caused a decrease in expansion ratio and an increase in bulk density. In contrast, hardness decreased, while fracturability increased with higher wholemeal spelt semolina content. The results of the colour analysis showed that the extruded samples were lighter and the total colour change (ΔE) increased proportionally with the addition of wholemeal spelt semolina.

Keywords: extrusion, wholemeal spelt semolina, corn snack products, physical properties

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Selective re-testing of promising herbicide active ingredients in lupine: final-year findings

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ABSTRACT

Sweet white lupine (*Lupinus albus* L.) is a high-protein legume with significant soil-improving potential through nitrogen fixation. However, its poor competitive ability against weeds often hinders cultivation. Our study aimed to select safe herbicides by testing various modes of action in the lupine. During the growing seasons of sweet white lupine, 15 active ingredients were tested at multiple doses, either alone or in combinations. In the final year, only those active ingredients that had proven suitable based on the preliminary trials were applied. Based on the final-year results, including NDVI, phytotoxicity, and seed yield data, the active ingredients flumioxazin (30 g ai ha⁻¹), pendimethalin (2275 g ai ha⁻¹), dimethenamid-P (1008 g ai ha⁻¹), pethoxamid (1200 g ai ha⁻¹), clomazone (95 g ai ha⁻¹), and metobromuron (1500 g ai ha⁻¹) are safe for use in pre-emergence technology. Regarding the active ingredient diflufenican (125 g ai ha⁻¹), one preliminary test year preceded our main experimental year; however, no significant differences were observed in the evaluated parameters compared to the control, and therefore, the results are considered promising. In addition to diflufenican, based on the NDVI, phytotoxicity, and seed yield results under post-emergence application, the active ingredients halauxifen-methyl (1.3 g ai ha⁻¹) and sulfosulfuron (9.8 g ai ha⁻¹) can also be recommended for further testing. In the case of flumioxazin, chlorotoluron (1400 g ai ha⁻¹), and carfentrazone-ethyl (14 g ai ha⁻¹), treated lupine plant height was significantly lower compared to the control. These active ingredients caused severe phytotoxicity, and based on comparison with previous years' results, we concluded that their use is not safe in the crop, under post-emergence application. Phytotoxicity values showed a strong negative correlation with NDVI, plant height, and seed yield. Conversely, seed yield showed a strong positive correlation with NDVI and plant height data.

Keywords: NDVI, phytotoxicity, herbicide, pre-emergence, post-emergence

Acknowledgment: Project C1771371 has been implemented with the support provided by the Ministry of Culture and Innovation of Hungary from the National Research, Development and Innovation Fund, financed under the KDP-2021 funding scheme.

Fungi as Causal Agents of Olive (*Olea europaea* L.) Decline in Herzegovina

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ABSTRACT

Olive (*Olea europaea* L.) is a very important crop grown in the Mediterranean part of Bosnia and Herzegovina. In the past decade, the number of olive trees has increased significantly. Like any plant species of agronomic interest, olive trees are subject to severe attacks from a variety of pathogens that affect their health and, ultimately, their yield and oil organoleptic quality. Dieback and wilting symptoms caused by phytopathogenic fungi are currently among the most serious diseases threatening olive trees in many countries of the Mediterranean basin. During May and September 2023 and 2024, field research was conducted with the aim of collecting samples of olive plant material infected by phytopathogenic fungi. A visual inspection of olive trees was carried out at 56 locations in the city of Mostar and the municipalities of Ljubuški, Čitluk, Neum, and Stolac. Based on the morphology of colonies in pure culture, as well as the appearance, size, and color of spores, fungi were identified to the genus level. Identification to the species level was performed using molecular methods based on polymerase chain reaction (PCR), sequencing, and sequence analysis. Pathogenicity tests were conducted in a greenhouse on branches of the four-year-old cultivar 'Oblica'. Based on the conducted research, the species *Diplodia seriata*, *Neocosmospora solani*, *Phoma* sp., and *Pythium* sp. were identified as causal agents of partial or complete dieback of olive trees.

Keywords: phytopathogenic fungi, olive dieback, *Olea europaea*, Herzegovina

Volatilome Shifts in Dual Culture: VOCs Interactions Between the Biocontrol Agent *Meyerozyma caribbica* and *Penicillium expansum*

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ABSTRACT

Blue mold, caused by *Penicillium expansum*, is one of the most important postharvest diseases, leading to significant economic losses and raising food safety concerns due to patulin contamination. The objective of this study was to examine volatilome changes during dual culture interactions between the biocontrol yeast *Meyerozyma caribbica* BBJ and the pathogen *P. expansum*, focusing on the role of volatile organic compounds (VOCs) in pathogen suppression. In dual culture assays, BBJ significantly inhibited fungal growth, reducing colony diameter by 55% by water-soluble compounds, while VOCs-mediated inhibition reached 47%. Headspace SPME–GC–MS analysis identified 43 VOCs from different chemical classes, showing distinct differences between monocultures and co-culture conditions. The co-culture volatilome was characterized by increased presence of compounds such as p-ethylguaiacol, phenol derivatives, and alcohols (1-propanol-2-methyl, 1-butanol-2-methyl), while several pathogen-associated volatiles, including farnesene and related terpenoids, were significantly reduced. These changes indicate that the presence of the biocontrol agent interferes with pathogen metabolism, potentially reducing its virulence.

The results indicate that VOCs represent an important component of the antagonistic activity of *M. caribbica*. In addition to direct inhibition of fungal growth, volatilome shifts reflect metabolic interactions between the two organisms that are relevant for disease development. This study provides a clearer insight into yeast–pathogen interactions and supports the further development of volatilome-based approaches for the postharvest disease management.

Keywords: volatile organic compounds, biocontrol, yeast, blue mold

Mono- and disaccharides as chemical markers of potato geographical origin: an analytical and chemometric approach

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ABSTRACT

Quantitative determination of primary metabolites in potato tubers is an analytical challenge due to the complexity of the matrix (high starch content) and the chemical diversity of the analytes, but it is crucial for the development of chemometric models of differentiation according to geographical origin and growing conditions. In this study, the emphasis was placed on mono- and disaccharides (glucose, fructose and sucrose) as potential chemical markers of origin. Tuber samples were collected in seven cycles during one growing season from five counties of the Republic of Croatia (Lika-Senj, Dubrovnik-Neretva, Istria, Međimurje and Požega-Slavonia counties). The mass fractions of mono- and disaccharides were determined by liquid chromatography with refractometric detection (LC-RID) according to a validated protocol, with confirmed linearity, selectivity and repeatability of the method. The obtained concentrations were within the expected physiological ranges for potato tubers: fructose 0.0769–1.0318 g 100 g⁻¹ (average 0.4736 g 100 g⁻¹), glucose 0.1610–2.0949 g 100 g⁻¹ (average 0.8204 g 100 g⁻¹) and sucrose 0.2090–1.7057 g 100 g⁻¹ (average 0.4699 g 100 g⁻¹). The results confirm the suitability of the HPLC-RI method for reliable quantification of the targeted mono- and disaccharides, which can be the basis for building a multi-analyte platform for authenticating the geographical origin of potatoes. In addition, the results show significant variability of concentrations depending on the location and time of sampling, confirming that these sugars are relevant chemical markers. This study lays the analytical foundation for multivariate data analysis (PCA and LDA), enabling high accuracy in classifying potatoes by county of cultivation, which is crucial for protecting product authenticity in the market.

Keywords: potato, geographical origin, primary metabolites, mono- and disaccharides, HPLC-RI, chemometrics, multivariate analysis.

Acknowledgment: This research was funded by the Croatian Science Foundation through the funding of the project, IP-2024-05-5900. This work was carried out within the project “Food Safety and Quality Center” (KK.01.1.1.02.0004). The project is co-financed by the European Union from the European Regional Development Fund.

Frequency of 1RS wheat-rye translocation in diverse wheat germplasm

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ABSTRACT

The 1RS translocation is one of the most widely utilized chromosomal rearrangements in wheat improvement, involving the transfer of the short arm of rye chromosome 1R onto wheat chromosomes. Among the identified wheat-rye translocations, the most well-known are 1AL/1RS and 1BL/1RS, in which the short arm of rye chromosome 1 (1RS) is transferred to wheat chromosomes 1A and 1B, respectively. The 1RS translocation is associated with increased abiotic and biotic stress tolerance, adaptability and yield components. However, its presence is often associated with reduced end-use quality, particularly in terms of bread-making properties. The aim of this research was to examine the frequency of 1RS translocation in a diverse set of 120 hexaploid wheat genotypes using molecular markers. Genomic DNA was extracted using modified CTAB protocol, and four pairs of primers were used for the PCR analysis (RYE-NOR, SEC A2/A3, PAW 161 and RIS). PCR products were analyzed using horizontal gel electrophoresis. The frequency of 1RS translocation ranged from 8% (RYE-NOR) to 19% (RIS) in analyzed wheat germplasm. Considering the origin of the genotypes, the highest frequency of the translocation was observed in genotypes originating from Eastern and Southern Europe. Regarding the year of release, the translocation was most frequent in genotypes released between 1980 and 2000. Understanding the prevalence of the 1RS wheat-rye translocation is important for the effective use of wheat germplasm in breeding programs, as it is associated with important agronomic traits of wheat.

Keywords: *Triticum aestivum* L., *Secale cereale* L., molecular markers, yield, quality

Preliminary investigation on the cultivation of *Physalis peruviana* in decoupled aquaponics

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ABSTRACT

The application of innovative agricultural technologies creates new opportunities. Aquaponics enables efficient crop production through the utilization of wastewater from intensive aquaculture systems. Due to its high nutrient, vitamin, and antioxidant content, physalis is a valuable food crop. The experiment was conducted in a double-walled plastic greenhouse using a decoupled aquaponics system, allowing the evaluation of plant responses without direct fish involvement. Thirty plants were assessed by measuring SPAD, NDVI, and yield parameters (total yield, berry weight, length, and diameter based on a sample of 10 fruits). Light intensity, air humidity, and temperature were continuously recorded. After a two-week establishment period following planting (April 23), SPAD and NDVI values were measured weekly (14 assessments). SPAD values ranged from 25.24 to 36.14, while NDVI ranged from 58.83 to 87.70 on average. Both indices increased rapidly, peaking by the end of July, followed by a gradual decline. Pearson's correlation analysis using environmental data from one and four weeks before harvest showed weak negative correlations between SPAD values and increasing temperature over four weeks. NDVI exhibited weak positive correlations with humidity and light intensity in both intervals. Harvest was carried out five times between July 9 and August 6, resulting in a total yield of 3936.80 kg ha⁻¹. Temperature and light intensity in the week preceding harvest showed moderate positive correlations with yield parameters, while four-week humidity data showed weak to moderate positive relationships. Overall, the aquaponics system provided suitable conditions for physalis cultivation, particularly where field production is limited. Optimizing environmental parameters may further improve yield stability.

Keywords: aquaponics, *Physalis*, cultivation, yield

Biocontrol potential of *Bacillus amyloliquefaciens* against *Pectobacterium*-associated potato soft rot

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ABSTRACT

Pectobacterium species represent a threat to the sustainability of potato production and storage worldwide. Control of these devastating pathogens became extremely difficult, considering the general lack of bactericides. Therefore, significant efforts are needed to find out efficient ways to control the tuber soft rot in storage conditions, as well as to prevent infections during the potato growing season in field. This study evaluates the biocontrol potential of the highly potent *B. amyloliquefaciens* strain ABO2 against *Pectobacterium*-associated potato soft rot. The antimicrobial efficacy of the strain was initially confirmed *in vitro* against *Pectobacterium* species originating from potato grown in Serbia, as well as reference strains from Lebanon and Brazil. Its performance was further assessed through a series of *in planta* bioassays. The effects of the treatment were evaluated using a potato slice assay, which enabled the assessment of tissue maceration in the susceptible potato cultivar VR808. Two experimental approaches were applied - simultaneous application of the biocontrol agent and pathogen inoculation to the center of wounded potato slices, and preventive treatment, involving the biocontrol agent application 24 h prior to pathogen inoculation. The results demonstrated that both preventive treatment and simultaneous application effectively suppressed disease development to levels comparable to the non-inoculated control. A tuber assay was additionally conducted by immersing wounded potato tubers in the cultivation broth of ABO2 strain 24 h prior to immersing in *Pectobacterium* spp. suspension, simulating natural tuber infection. This treatment as well resulted in inhibition of tuber soft rot development, with disease severity reduced to levels observed in the non-inoculated control. These results indicate that *B. amyloliquefaciens* ABO2 is a promising candidate for sustainable management of potato soft rot through preventive tuber treatment prior to storage.

Keywords: soft rot, *Pectobacterium*, biocontrol, *Bacillus amyloliquefaciens*

Biofortification with Se and Zn - an integrated approach to improving the nutritional value of crops and human health

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ABSTRACT

Enrichment of crops with selenium (Se) and zinc (Zn) is an increasingly important strategy for improving the nutritional quality of agricultural products and supporting population health. These essential micronutrients, although required in trace amounts, are vital for human health, particularly in immune function, antioxidant defense, and metabolic regulation. However, many soils worldwide, including those in Europe and Croatia, show low natural availability, especially of Se, which is directly reflected in the content in food products of plant origin. This paper aims to synthesize recent findings from agronomy, food science and nutrition and promote interdisciplinary approaches to crop biofortification. Special emphasis was placed on nutritional implications, specifically how increasing the content of these micronutrients in crops may prevent dietary deficiencies, improve nutritional quality and contribute to long-term health outcomes. In this paper, the average daily intake of Zn and Se was estimated across eight scenarios (four omnivorous cases and four vegetarian cases). Based on USDA food composition data, adequate intake levels were observed for both men and women in all scenarios. Different scenarios were also compared with menus prepared using ChatGPT and menus developed by professional nutritionists, and in all cases, Zn and Se intake met recommended levels according to USDA data. However, when nutrient composition data specific to food produced and/or consumed in Croatia were applied, notable differences emerged. Modeled Zn intake remained comparable (99.4-102.3% of USDA based estimates), whereas Se intake was 15.2-31% lower. Although Se intake remained within adequate ranges in these optimized dietary models, it is important to note that these scenarios do not reflect actual dietary habits in Croatia. Therefore, the study highlights the importance of assessing actual generally dietary intake and the intake of Zn and Se at the national level. In Croatia, we can expect significantly lower, perhaps even too low intakes of Zn and especially Se depending on dietary habits and actual concentrations of Zn and Se in food on the Croatian market, indirectly increasing the health-risk.

Keywords: selenium, zinc, malnutrition, soil conditioning, nutritional profile, health-risk

Quality characteristics of muffins made with zinc and selenium-biofortified barley flour

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ABSTRACT

Barley flour offers various nutritional benefits compared to wheat flour, but reduced gluten functionality can affect baked product quality. This study evaluated the quality of muffins formulated with zinc- and selenium-biofortified barley flour (one hulled and one hulless biofortified barley variety). Four formulations were prepared: 100% wheat flour (control), 50/50% barley/wheat flour, 100% barley flour, and 80/20% barley flour/vital gluten muffins. Two types of barley flour were used: integral (from hulled and hulless barley) and refined (pearled hulled and hulless flour). Muffin quality was evaluated by measuring specific volume, porosity, textural profile analysis (TPA), color using the CIELab system, and sensory analysis. Formulations containing wheat flour exhibited higher specific volume, lower crumb hardness, and greater sensory acceptance than 100% barley flour muffins. Barley inclusion produced a darker color and greater heterogeneity in porosity. However, muffins with 50/50% barley/wheat flour and 80/20% barley flour/vital gluten showed acceptable quality characteristics.

Nutritional analysis confirmed significantly higher concentrations of selenium and zinc, as well as an increased proportion of β -glucan in muffins made from barley flour compared to wheat samples. Overall, the results indicate that, using biofortified barley, acceptable muffins with improved nutritional value and functional potential can be produced through appropriate recipe adjustments.

Keywords: barley flour, muffins, zinc, selenium, biofortification, functional bakery products

Are there genotypic differences in the success of agronomic biofortification of barley with selenium and zinc?

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ABSTRACT

Insufficient concentration of microelements in the soil and/or edible parts of crops is an increasingly common cause of malnutrition in people worldwide and in Croatia. Therefore, a field experiment was conducted on eutric brown soil in the Osijek area in the 2024/25 vegetation season to investigate whether there is genetic specificity of barley cultivars in terms of Se and Zn accumulation in grain (research of 8 barley cultivars, different varieties), and whether agronomic and microbiological biofortification methods can be successfully applied in barley cultivation. Agronomic biofortification was carried out at the beginning of barley flowering by foliar application of Se (10 g ha⁻¹) or Zn (3 kg ha⁻¹). Microbiological biofortification was carried out by treating barley seeds immediately before sowing with a mixture of microorganisms. The average grain yield was 8,323 kg ha⁻¹ without significant influence of treatments, although the highest yield was achieved with the application of microorganisms or with the application of Se, and the lowest with the application of Zn, but the average differences of 200-368 kg ha⁻¹ were not statistically significant. Foliar application of Zn resulted in an average increase in Zn concentration in grain of 130% (48.15 vs. 20.97 mg kg⁻¹), and Se of 19.5 times (540.0 vs. 27.7 µg kg⁻¹). It is very significant that all cultivars achieved a target concentration of Zn > 40 mg kg⁻¹ (highest in the cultivars Mandatar 56.14 and Otkos 51.54 mg kg⁻¹) and Se > 300 µg kg⁻¹ (highest in the cultivars Osvit 663.7 and Mandatar 619.5 µg kg⁻¹). On average, biofortification with microorganisms did not significantly increase the concentration of Zn and Se, although the reaction of the genotypes was different. In conclusion, not just that the successful biofortification of all barley genotypes with Zn and Se above target concentration was achieved, but also the genotypic diversity in Zn accumulations in all treatments and Se accumulations if Se was applied.

Keywords: yield, foliar application, two-row barley, multi-row barley, hullless barley

Designing Botanical Spirits Using Agro-Industrial By-Products and Tea-Derived Materials

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ABSTRACT

The formulation of botanical spirits is increasingly shifting from an intuitive craft practice towards a structured, science-informed process. In this context, agro-industrial by-products and underutilized plant materials, including citrus peels, fruit pomace, spent tea leaves, herbal residues, and floral processing fractions, offer a promising source of sustainable botanicals for distinctive distilled beverages. These materials often retain substantial levels of volatile and semi-volatile compounds that can impart citrus, floral, herbal, spicy, woody, and earthy sensory characteristics. However, their effective use requires careful control of raw material selection, geographical origin, pre-treatment, particle size, solvent composition, and distillation parameters. The extraction of aroma-active compounds is strongly dependent on the ethanol-water ratio, as target molecules vary in polarity, solubility, and volatility. This study will present recommendations for optimising botanical extraction from tea-derived materials and agro-industrial by-products intended for use in distilled beverages. Emphasis will be placed on defining suitable alcohol strength according to the polarity and sensory profile of target compounds, as well as on optimising maceration time, particle size reduction, and controlled heating to maximise extraction efficiency and preserve desirable aroma quality. The study will further address distillation fractionation as a critical step for retaining volatile top notes while limiting the transfer of heavier fractions associated with less desirable sensory impressions. Through this approach, the study will highlight practical strategies for the valorisation of low-value aromatic side streams and their transformation into functional botanicals. These recommendations are expected to support product differentiation and more sustainable flavour design, while also contributing to raw material efficiency and circular bioeconomy objectives in modern distilled beverage production.

Keywords: botanical spirits; agro-industrial by-products; tea residues; flavor extraction; circular bioeconomy

Acknowledgement: This research was conducted as part of the project „From food industry by-products to new functional products (NUS-PRO-FUN, 581-UNIOS-94)” funded by National Recovery and Resilience Plan (funded by the European Union, NextGenerationEU)

Zinc localization in *Triticum aestivum* L. grains and flour zinc content under ZnSO₄ fertilization

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ABSTRACT

Deficiencies in micronutrients, particularly zinc (Zn), affect a substantial portion of the global population, leading to weakened immune system and impaired growth, among others. Nevertheless, in staple crops such as *T. aestivum* L., Zn also plays an essential role, contributing to structural, functional and regulatory processes. This study aimed to compare zinc content in whole wheat flours with refined flours, while also determining its distribution within the grain, when applying ZnSO₄ fertilizer. In the experimental field in Beja (Portugal), a workflow for Zn enrichment by foliar application of ZnSO₄ in *T. aestivum* L. (cvs Roxo and Paiva) was implemented. Post-harvest analyses were conducted (e.g., ash content and zinc quantification by XRF in flours and zinc localization by micro-energy dispersive X-ray fluorescence system in grains) after two foliar applications of ZnSO₄. Foliar applications took place at booting and heading stages in three concentrations (P0 and R0 – 0 (control); P1 and R1 – 8.1 and P2 and R2 – 18.2 kg ha⁻¹). The results indicated that Zn was predominantly localized in the embryo and in the vascular bundle of the grains. Zinc application also incremented Zn concentration in both grains and flours, with whole wheat flours exhibiting higher levels than refined flours. To sum up, these findings support that whole wheat flours offer a superior nutritional product, enabling the development of innovative and functional food products enriched in Zn.

Keywords: bread wheat flours, *Triticum aestivum* L. grains, Zn quantification, ZnSO₄ fertilization

Kinetics of P, K and Ca uptake, translocation and accumulation in *Triticum durum* Desf.

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ABSTRACT

This study evaluated the effects of three biostimulants (BlueN® - T6 and T7, Contribute ibNP – T2 and T3, and Status® - T4 and T5) combined with standard (T1, T2, T4 and T6) and reduced (T3, T5 and T7) fertilization regimes (Foskamónio 12-24-12, 150 – 113 kg ha⁻¹) on the dynamics of P, K, and Ca uptake, transport, and accumulation in durum wheat (*Triticum durum* Desf.). Dry matter allocation revealed a treatment-driven modulation of source–sink dynamics, with T6 enhancing vegetative biomass and T7 promoting a stronger allocation towards grains, indicative of increased sink strength during grain filling. Macronutrient accumulation reflected coordinated regulation between root uptake capacity and long-distance transport. Total P, K, and Ca accumulation per plant reached maxima under T7 (13.90 mg, 170.55 mg, and 112.60 mg, respectively), suggesting enhanced acquisition efficiency and vascular loading. K exhibited the highest mobility and grain allocation, but Ca revealed constrained redistribution, consistent with a limited phloem mobility. P displayed intermediate behavior, with effective remobilization to reproductive tissues. Grain nutrient loading was therefore governed by differential element mobility and treatment-induced shifts in transport efficiency. These findings support a mechanistic linkage between biomass production, vascular transport processes, and nutrient partitioning, highlighting the role of sink-driven regulation in optimizing nutrient use efficiency.

Keywords: biostimulants, macroelements accumulation, macroelements translocation, *Triticum durum* Desf.

OLIVE – EduTech: An interdisciplinary approach to monitoring olive tree response to pruning

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ABSTRACT

Pruning olive is a key agrotechnical measure that regulates yield and facilitates the application of other cultivation practices. Despite the well-known importance of pruning, the number of scientific publications on this topic is limited, and there are different views among experts about its impact on yield. Therefore, an interdisciplinary research group (agronomy, geography, pedagogy, and chemistry) will be formed within the OLIVE – EduTech project, with the aim of determining the physiological response of olive trees to different pruning intensities and developing methods for mediating the transfer of knowledge to olive growers who apply this practice to a limited extent. The specific objective of the project is to quantify the impact of different pruning intensities on yield, olive physiology, fruit quality and the composition of olive oil, minerals, and phenols in the leaf. The impact of pruning will be quantified using spectral and morphometric parameters, by estimating the amount of removed biomass, and by chemical analyses. In addition, students and olive growers will be educated on the importance of proper pruning, and the acquisition of knowledge about this skill will be monitored. Expected results include improving knowledge about olive pruning, introducing new methods and developing specialized equipment for fruit and olive oil analysis at the University of Zadar, increasing scientific productivity, and popularizing science in the field of olive growing.

Keywords: education, geospatial technologies, yield, olive leaf, olive fruit, workshop

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Sunflower hybrid responses to plant density: Linking morphology and photosynthetic efficiency

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ABSTRACT

This study investigated the effects of three planting densities (84034, 68027, and 57143 plants/ha) on agronomic and physiological properties of five sunflower hybrids (Surimi CL, Integral CL, Alexa SU, Neta SU, and Davero SU) in a field experiment. Agronomic properties such as plant height, stem and head diameter, number of leaves, grain yield, oil content, and 1000-grain weight were measured. Physiological properties were assessed using chlorophyll *a* fluorescence at three developmental stages: budding, flowering, and grain-filling. Analysis of variance revealed that plant density had a significant impact on most agronomic properties, except leaf number and hectoliter mass. Hybrid differences were significant for all properties except head diameter. Significant interactions (treatment × hybrid) were found only for grain yield, oil content, and oil yield. Denser plantings generally led to taller plants but smaller stem and head diameters, while lower densities favoured greater 1000-grain weight and hectoliter mass in some hybrids. Photosynthetic parameters varied markedly across developmental stages, with higher ABS/RC, TR_o/RC, and DI_o/RC values observed at the budding stage. Correlation analysis revealed trade-offs between structural growth and photosynthetic efficiency, especially during early reproductive stages. Genotype-specific responses indicated that Alexa SU and Neta SU were more sensitive to density changes, while Integral CL and Surimi CL maintained greater physiological stability. These findings underscore the potential of integrating physiological and agronomic properties to guide hybrid-specific planting density optimisation under varying environmental conditions.

Keywords: hybrid, agronomic traits, chlorophyll fluorescence, environmental conditions

Effects of elevated CO₂ on starlet and normal flowers of *Coffea arabica* L.

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ABSTRACT

Climate changes and global warming are expected to impact plant growth and reproduction. Notably, the concomitant rise in air [CO₂] can mitigate ecophysiological drought and heat impacts, but the potential effects on floral developmental stability remain poorly understood in perennial crops, such as *Coffea arabica* L. This study aimed to compare the occurrence and morphological traits of abnormal (starlet) and normal flowers in four year old potted *C. arabica* cv. Icatu plants, grown under similar environmental conditions, but differing in ambient (aCO₂, 420 ppm) or elevated (eCO₂, 700 ppm) [CO₂] in air, to assess the role of eCO₂ on floral phenotype expression (flower morphology and the frequency of starlet and normal flowers). The number of starlet flowers significantly declined in eCO₂ than in aCO₂, with eCO₂ promoting greater floral developmental stability, with a lower incidence of starlet phenotypes and more consistent floral structures. This suggests that increased C-availability (due to greater leaf photosynthetic rates) may mitigate factors underlying abnormal floral development, possibly through enhanced physiological balance during reproductive organ formation. Overall, eCO₂ appears to exert a mitigating effect on floral abnormalities in *C. arabica* as compared with ambient conditions, which may partially buffer reproductive instability, emphasizing the importance of considering floral developmental responses when predicting coffee crop performance under future climate scenarios.

Keywords: *Coffea arabica*, floral morphology, elevated CO₂, reproductive development, phenotypic variation

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Drought constraints and air CO₂ enrichment: Implications for photosynthesis in elite genotypes of *Coffea arabica* L.

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ABSTRACT

Ongoing climate changes are intensifying drought frequency and severity, threatening the sustainability of *Coffea arabica* L. crop. Elevated atmospheric [CO₂] (eCO₂) may partially offset these impacts by enhancing C-assimilation, although its role under water deficit should be better understood. Photosynthetic performance was assessed in newly matured leaves of plants from *C. arabica* cvs. Geisha 3 (G3), Marsellesa (Mar), and their Hybrid (Hy), grown under well-watered conditions (control, WW) and either normal (400 μL L⁻¹, aCO₂) or elevated (700 μL L⁻¹, eCO₂) atmospheric [CO₂]. Afterwards, plants were submitted to moderate (MWD) and severe (SWD) drought by partially withholding irrigation for *ca.* 20 days, reaching predawn leaf water potentials of *ca.* > -0.4 MPa, -1.2 to -1.8 MPa, and -2.6 to -3.0 MPa, respectively. Subsequently, irrigation was resumed, and plant recovery was monitored over a period up to 14 days (Rec14). The decline in water availability gradually reduced net photosynthetic rate (P_n) and stomatal conductance (g_s) in all genotypes, but P_n values remained significantly 2-3x higher under eCO₂ than aCO₂ (G3: WW +39%, MWD: +26%, SWD +213%; Mar: +59%, +79%, +15%; and Hy: +58%, +70%, +250%). Under aCO₂, increased C_i under SWD indicated predominant non-stomatal limitations. Initial and total RuBisCO activity generally decreased under MWD and SWD (*vs.* WW) at both [CO₂] levels, although G3 showed slightly higher activity under SWD compared with MWD. eCO₂ mitigated drought effects, enhancing P_n across water treatments. Furthermore, all genotypes showed only a partial recovery of P_n and g_s by Rec14, despite an almost full recovery for both RuBisCO activities, with a slightly better recovery under eCO₂. Overall, G3 displayed greater drought resilience and recovery, whereas Mar seems to display greater sensitivity. These findings highlight the mitigating effect of eCO₂ on coffee photosynthesis, with genotype-dependent RuBisCO responses under future climate scenarios.

Keywords: *Coffea arabica* L., Drought, Elevated CO₂, Photosynthesis, RuBisCO.

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Impact of prefermentative ultrasound treatments on the concentrations of macro- and microelements in Malvazija istarska wines

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ABSTRACT

The mineral composition of wine significantly affects its quality and nutritional value. Since mineral concentrations may be influenced by technological practices during vinification, this study aimed to investigate the impact of prefermentative ultrasound treatments with different amplitudes and durations on the mineral profile of Malvazija istarska wines. During the 2024 harvest, six different vinification treatments were carried out. Four treatments were based on the application of a prefermentative ultrasound technique on cooled cryomacerated mash (10 °C) as follows: ultrasound treatments of 70% amplitude for 80 minutes (US80-70%) and 160 minutes (US160-70%), and ultrasound treatments of 100% amplitude for the same durations as the previous ones (US80-100% and US160-100%). In addition to these treatments, the research also included a control treatment – C (wine produced using standard white winemaking technology) and a cryomaceration treatment lasting one day at 10 °C (CRIO). The research was conducted using an industrial ultrasonic processor, UIP2000hdT-230 (20 kHz, 2000 W). Concentrations of macro- and microelements were determined by ICP-OES. Nine elements (Ca, K, Mg, Na, P, S, B, Li and Se) were found in the analyzed wines. Potassium (K) was the most abundant element in all wines, with ultrasound treatments showing significantly higher concentrations compared to the CRIO and C treatments. The highest concentrations of Mg, Na, B, P, and S were observed in the US160-100% treatment, while the C wine showed the lowest concentrations. Elements such as Al, Cd, Co, Cr, Cu, Fe, Mn, Mo, Ni, Pb, Si and Zn were not detected in the analyzed wines. The highest concentrations of total macroelements, as well as the sum of all detected elements, were observed in the US160-100% treatment, while the lowest concentrations were found in the control wine. These results indicate that higher amplitudes and longer durations of ultrasound enhance the extraction of mineral elements from grape solids into wine, significantly modifying its mineral profile compared to conventional vinification techniques.

Keywords: ultrasound treatments, prefermentative treatments, macroelements, microelements, Malvazija istarska wines

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Distinct photochemical and recovery-driven strategies underlie genotype-specific cold priming responses in maize

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ABSTRACT

Cold stress during early seedling development is a major constraint on maize (*Zea mays* L.) production in temperate regions. This study evaluated effects of a graduated cold priming protocol on six maize inbred lines. Three treatments were applied: optimal conditions, cold stress, and cold stress preceded by graduated chilling priming. Chlorophyll *a* fluorescence (OJIP) was recorded at five time points across stress and recovery; biochemical analyses at peak stress and at the end of recovery. Primed plants showed significantly higher maximum quantum efficiency of photosystem II photochemistry (F_v/F_m) at peak stress than non-primed stressed plants (0.601 vs 0.506; $p = 0.017$), and lower lipid peroxidation, with MDA not differing from controls ($p = 0.507$). Recovery of F_v/F_m to $\geq 95\%$ of baseline was achieved by all primed genotypes within 72 h, versus only one of six non-primed genotypes. Total chlorophyll recovered to control levels in primed plants by the final recovery point, while non-primed plants remained significantly depleted ($p < 0.001$). Final dry biomass was lower in primed than cold-stressed plants (0.39 g vs 0.56 g; $p = 0.008$), reflecting greater cumulative cold load. Dry matter content did not differ from controls (7.81% vs 7.92%; $p = 0.649$), indicating preserved assimilate allocation rather than qualitative tissue impairment. Genotypic responsiveness varied substantially: Mo17 showed the highest Composite Priming Benefit Score (0.89) and Os6251/15 the lowest (0.30). Graduated cold priming effectively reduced photosystem II damage and accelerated multi-trait physiological recovery, with benefit magnitude being strongly genotype-dependent. Intermittent cold episodes common in temperate spring climates may act as natural priming events, enhancing maize seedling capacity to withstand subsequent cold stress.

Keywords: maize, cold priming, cold stress, recovery, genotype-dependent response

Antagonistic Potential of *Trichoderma* spp. Against *Alternaria* Apple Rot

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ABSTRACT

Apple production is a vital agricultural sector in Serbia, yet postharvest storage is frequently compromised by fruit rot caused by *Alternaria* spp. With increasing pathogen resistance to synthetic fungicides and stricter regulations on chemical residues, there is a pressing need for sustainable biological control strategies. The objective of this study was to isolate and evaluate the antagonistic potential of autochthonous *Trichoderma* spp. strains obtained from the rhizosphere of apple trees at the Bački Jarak locality. Soil samples were collected from a depth of 0.10 to 0.15 m. Isolation was performed using the serial dilution method on selective nutrient media supplemented with antibiotics to inhibit bacterial growth, followed by incubation at 25 °C. Based on macroscopic colony characteristics and microscopic analysis of reproductive structures, seven distinct *Trichoderma* isolates were identified and purified. Their biocontrol efficacy against *Alternaria* spp. was assessed using the *in vitro* dual culture method on potato dextrose agar, with mycelial discs placed at a distance of 3 cm. After incubation for seven days at 25 ± 2 °C, all tested isolates demonstrated significant antagonistic activity. The growth inhibition percentages of the pathogen were high, ranging from 78.9% to 89.4%. These results indicate that local soil-borne isolates possess a potent fungicidal effect. In conclusion, the identified *Trichoderma* strains are promising candidates for the development of biofungicides. Future research should focus on molecular identification of the species and *in vivo* application on stored fruits to validate their protective capability under real storage conditions.

Keywords: *Malus domestica*, biological control, *Trichoderma* spp.

Oxidative Stress and Antioxidant Response of Sour Cherry (*Prunus cerasus* L.) Varieties under Heat Stress

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ABSTRACT

Sour cherry (*Prunus cerasus* L.) is a commercially important fruit species with high antioxidant and nutritional value and one of the most widely used fruits in the food industry. However, research on its responses to abiotic stress remains limited, despite increasingly harsh conditions caused by global climate change. The aim of this study was to evaluate differences in the response of two sour cherry cultivars, 'Érdy jubileum' and 'Debreceni bötermő' under heat stress condition. One-year old potted plants were exposed to 25 °C (control) and 42 °C (heat stress) under controlled growth chamber conditions. Heat stress induced significant differences in oxidative and antioxidative responses between cultivars. 'Érdy jubileum' showed higher H₂O₂ content (1.122 μmol H₂O₂ g⁻¹ FW) compared with 'Debreceni bötermő' (0.6459 μmol H₂O₂ g⁻¹ FW), indicating more efficient ROS regulation in 'Debreceni bötermő'. Higher activity of catalase (CAT) and ascorbate peroxidase (APX) was found only in 'Érdy jubileum' (15.18 and 5.272 U mg⁻¹ protein, respectively). In contrast, guaiacol peroxidase (GPOD) along with polyphenol oxidase (PPO) showed stronger activity in 'Debreceni bötermő' (0.63 and 3.31 U mg⁻¹ protein). The contents of total polyphenols (13.676 mg GAE g⁻¹ FW) and flavonoids (7.82 mg QE g⁻¹ FW) as well as the antioxidant capacity using ABTS method was higher in 'Debreceni bötermő' (56.70 mmol TE g⁻¹ FW) than in 'Érdy jubileum' (11.011 mg GAE g⁻¹ FW, 6.17 mg QE g⁻¹ FW, and 41.76 mmol TE g⁻¹ FW, respectively), this was also confirmed using FRAP metode. Overall, the results indicate that 'Debreceni bötermő' exhibits greater tolerance to heat stress, which was confirmed by lower oxidative stress and higher antioxidant capacity compared with 'Érdy jubileum'.

Keywords: sour cherry, heat stress, antioxidant response, ROS, polyphenols

Trace Element Profile in Potato Tubers as a Basis for Multivariate Assessment of Geographical Origin

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ABSTRACT

The mineral composition of potato tubers is one of the key physicochemical characteristics that can serve as a basis for developing models for determining geographical origin and cultivation systems. While essential elements (Cu, Fe, Mn, Zn, Na, Ni, and Cr) play an important biological role in metabolic processes, potentially toxic elements (Cd and Pb) have no nutritional value and may pose a risk to human health. The aim of this study was to determine the trace element profile in potato tubers from five counties of the Republic of Croatia and to assess their compliance with current legislative regulations. A total of 97 potato tuber samples were analyzed, collected during seven sampling periods from the areas of Lika-Senj, Dubrovnik-Neretva, Istria, Međimurje, and Požega-Slavonia counties. Samples were prepared using microwave digestion with nitric acid, after which the mineral composition was determined using inductively coupled plasma mass spectrometry (ICP-MS). Quantification was performed using the external standard method with method validation. The average concentrations of essential elements (expressed on a dry matter basis) were: Na 5.64 mg kg⁻¹, Fe 26.16 mg kg⁻¹, Mn 7.68 mg kg⁻¹ and Zn 46.98 mg kg⁻¹. A pronounced variability among samples was observed. For potentially toxic elements, the following average values were determined: Cd 0.26 mg kg⁻¹, Cr 0.11 mg kg⁻¹, Ni 1.36 mg kg⁻¹ and Pb 0.05 mg kg⁻¹. After recalculation to fresh weight (average dry matter content ≈19%), the concentrations of Cd and Pb in all samples were below the maximum permitted levels established by Regulation (EC) No.1881/2006. Maximum permitted levels for Cr and Ni in potatoes have not been established; however, the obtained values are consistent with ranges reported in available scientific literature. The results confirm the safety of the analyzed samples for human consumption and indicate the potential use of the mineral profile as a database for developing multivariate models for determining geographical origin and distinguishing potato cultivation systems.

Keywords: potato, ICP-MS, trace elements, geographical origin, food safety

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Valorization of Orange Peel Powders as Natural Stabilizers in Oil-in-Water Pickering Emulsions

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ABSTRACT

Orange peel (OP) is a major by-product of the citrus industry, accounting for approximately 20% of total citrus waste. Although commonly discarded, OP is rich in dietary fiber and bioactive compounds, offering significant opportunities for sustainable valorization. This study investigated the use of OP powders as natural stabilizers in oil-in-water Pickering emulsions, when micronized via advanced wet friction milling (AWFM). This novel technique enabled the production of sufficiently fine particles to effectively adsorb at the oil–water interface, a key requirement for Pickering stabilization. Upon homogenization, micronized citrus fibers formed hydrated particles capable of accumulating at the droplet interface and forming a protective barrier through a combination of physical and chemical stabilization mechanisms. OP powders were used either alone or in combination with bacterial cellulose (BC) as a natural stabilizing system. Emulsions were prepared using OP concentrations of 1.5% and 2.5% (w/w) and BC concentrations of 0.5% and 0.8% (w/w), while the oil phase was maintained at 4% (w/w). The stabilizers were characterized in terms of particle size, oil sorption capacity, and water and oil holding capacities. The produced Pickering emulsions exhibited good physical stability, with droplet size distributions characterized by $d_{0.9}$ values below 184.97 μm . Emulsion functionality was evaluated through spectrophotometric analyses, including total phenolic content, total flavonoid content, DPPH, and FRAP, along with physical stability over 28 days. Bioactivity measurements showed an increase up to 15 days, particularly in emulsions containing the highest OP concentration, followed by a slight decline thereafter. Overall, the findings demonstrate the effectiveness of OP powders as natural Pickering stabilizers. Furthermore, the combination of OP with BC improved the retention and potentially the controlled release of phenolic compounds and flavonoids, enhancing antioxidant activity.

Keywords: orange peel (OP), advanced wet friction milling (AWFM), Pickering emulsion, stabilizer

The effects of seed priming with sodium hydrosulphide on drought tolerance of sunflower (*Helianthus annuus* L.) seedlings

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ABSTRACT

This study investigated the effects of pre-sowing treatment of Luka sunflower hybrid seeds with water and sodium hydrosulfide (NaHS) under drought stress. The results indicate that the imposed water deficit, induced by the use of PEG under laboratory conditions or reduced soil capacity in outdoor conditions, drastically reduces germination energy, emergence rate, and fresh seedling mass accumulation. The metabolic response to oxidative stress was manifested through a significant accumulation of proline and malondialdehyde (MDA), confirming the disruption of cellular homeostasis under the influence of drought.

The application of NaHS during the germination showed promising but specific results. The positive effects on germination were strictly dependent on the concentration of the exogenous hydrogen sulfide donor and the level of stress, suggesting the existence of a limited concentration range within which NaHS exerts a stimulatory effect. However, as plants transitioned into the soil emergence phase, the initial advantages of chemical priming began to fade. Although drought activated a robust enzymatic antioxidant system in the leaves (CAT, APX, GR), the influence of NaHS remained largely limited to the modulation of DHAR enzyme activity.

In conclusion, the study indicates that NaHS treatment can provide initial support to seeds during germination under unfavorable conditions. Treatment with NaHS does not provide a long-lasting priming effect on the overall antioxidant status of the plant during later stages of early growth. Most of the recorded stimulatory effects in the soil can be attributed to the hydropriming process itself, while NaHS does not achieve a significant prolonged impact on the defense mechanisms of sunflower. These results suggest a need for further research into the stability of hydrogen sulfide signaling molecules in seed tissue to optimize application methods in agricultural practice.

Keywords: antioxidative enzymes, drought stress, NaHS, proline, seed priming, sunflower

Efficiency of Different Traps and Attractants for Mass Trapping of the Mediterranean Fruit Fly (*Ceratitis capitata* Wied.) in Herzegovina

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ABSTRACT

Monitoring and control of the Mediterranean fruit fly are carried out using several types of traps that differ in attractant type, capture method, and purpose (monitoring or mass trapping). The aim of this study was to determine which trap type, together with its corresponding attractant, most effectively reduces the population of the Mediterranean fruit fly in the Herzegovina region. The study was conducted in 2025 in a small family-owned citrus orchard at the Blizanci site in the municipality of Čitluk. The trial included visual traps (yellow sticky plates), visual traps with a food attractant (Csalomon PALz traps), traps with a sexual pheromone (Jackson traps), traps with food attractants (Csalomon RAG and Csalomon VARs traps), and traps used in the attract-and-kill method (Karate Trap C). Each trap type was tested in two replicates. The traps were installed on September 1 and inspected weekly until mid-November, when no further captures were recorded. One week after installation (September 7), moderate catches were recorded across all trap types. By the second week (September 14), a marked increase in population abundance was observed, particularly in Karate Trap C traps (89/102), indicating the onset of a more intensive autumn flight. Peak flight activity was recorded between September 21 and October 5, with the highest captures observed in Karate Trap C and Csalomon PALz traps. After mid-October, captures declined steadily, and by mid-November, they had ceased completely. A total of 3,678 individuals were captured during the study period. The highest efficiency was recorded for Karate Trap C (1,723 individuals), followed by Csalomon PALz and Csalomon VARs traps. Csalomon RAG and Jackson traps showed moderate capture rates, while yellow sticky plates were the least effective, with only 43 individuals captured.

Keywords: Mediterranean fruit fly, trap efficiency, Herzegovina

Protein Hydrolysate Application Modulates Phenolic Composition of Extra Virgin Olive Oil

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ABSTRACT

The aim of this study was to assess the impact of foliar-applied protein hydrolysates on olive oil quality and phenolic profile, with emphasis on secoiridoid compounds. The experiment was conducted in Zadar County (Škabrnja) as a completely randomized design on 12 olive trees of the 'Leccino' cultivar. Foliar treatments were applied four times at 10-day intervals, starting 50 days after anthesis. In addition to the untreated control, protein hydrolysate treatments were applied. Proteins were extracted from pumpkin seed cake and enzymatically hydrolyzed using Alcalase® Pure + Flavourzyme® (H2 treatment) or Alcalase® Pure + Protana™ Prime® (H3 treatment). No differences were observed in basic oil quality parameters, and all samples were classified as extra virgin olive oil. Statistical analysis revealed treatment-dependent differences in the phenolic composition of olive oil. Oleuropein aglycone showed higher values in the control compared to protein hydrolysate treatments, while flavonoids luteolin and apigenin exhibited the opposite trend, with the highest values in H3. Oleocanthal did not differ significantly among treatments, whereas oleacein was significantly higher in the control compared to H2, while H3 did not differ from the other treatments. Overall, protein hydrolysate treatments modulated the phenolic profile without compromising oil quality. Changes in oleuropein aglycone and oleacein indicate an effect on the secoiridoid pathway, more pronounced at the aglycone level. Given their role in bitterness and pungency, future research will focus on linking these changes with sensory properties and consumer acceptance.

Keywords: hydrolyzed proteins, pumpkin seed cake, biostimulants, secoiridoids, foliar application.

Effects of three biostimulants on the grain quality of *Triticum durum* Desf.

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ABSTRACT

Durum wheat (*Triticum durum* Desf.) is one of the most widely consumed cereals in the world. Its production is on the rise to keep pace with the growing global demand for staple foods, in a context that calls for the adoption of more sustainable agricultural practices. During the 2024/25 growing season, a field trial was established in Beja (Portugal) with one control plot and three treatment plots, each assigned a different biostimulant (Contribute ibNP, Status®, and Blue N®), combined with a standard fertilization regime (Foskamónio 12-24-12, 150 kg ha⁻¹). In this context, the present study aimed to evaluate the effects of application of these three biostimulants on the quality of durum wheat cv. Don Ricardo. To this end, the levels of nitrogen, total lipids, wet gluten, dry gluten, ash, moisture, and total sugars in the semolina produced from the durum wheat grain were quantified. The application of these biostimulants demonstrated an increased effect on total lipid, moisture, and ash content, whereas in the quantification of total sugars, only the Status® biostimulant yielded insufficient results. However, the values for nitrogen and gluten content (wet and dry) were lower than those observed in the control, which showed a higher yield. When comparing the three biostimulants tested, the Blue N® product stood out overall for its more favorable results.

Keywords: biostimulants, durum wheat, grain quality

Marker-assisted selection in alfalfa (*Medicago sativa* L.) breeding: Achievements, challenges and future perspectives

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ABSTRACT

Alfalfa (*Medicago sativa* L.) is one of the most important forage legumes worldwide due to its high nutritional value and significant role in sustainable agriculture. However, its cultivation is challenging due to its autotetraploid nature, allogamous reproductive system, pronounced heterozygosity, and other specificities of this crop. Traditional alfalfa breeding programs rely mostly on phenotypic recurrent selection, which is proven to be effective, but time-consuming and strongly influenced by genotype-environment interactions. To overcome these limitations, modern programs should incorporate molecular tools such as marker-assisted selection (MAS), enabling the identification and selection of genotypes carrying desirable alleles for traits including biomass yield, forage quality, and resistance to biotic and abiotic stresses. This allows selection at early developmental stages, before field testing, independently of environmental effects, potentially shortening the breeding cycle and increasing selection accuracy. Despite its great potential, the practical application of MAS in alfalfa breeding programs remains limited, as most research to date has focused on identifying molecular markers and genomic regions, rather than validating whether selected genotypes actually improve field performance, which is partly due to the long breeding cycles of alfalfa. The slow and labor-intensive collection of accurate phenotypic data under field conditions is a major bottleneck, which can be addressed by high-throughput phenotyping using handheld sensors and aerial or satellite remote sensing combined with machine learning, complementing MAS by more efficiently linking genotype and phenotype. Broader implementation of MAS will require development of molecular technologies, stronger collaboration between researchers and breeders, and investment in education and digital tools. Together, these approaches can accelerate breeding, enhance selection precision, and support the development of more resilient and productive alfalfa cultivars.

Keywords: forage legumes, breeding, molecular tools, marker-assisted selection, traits

Acknowledgement: This research was funded by the National Recovery and Resilience Plan (NRRP) 2021-2026 (C3.2. R1-11) through the Program Agreement between the Agricultural Institute Osijek and the Croatian Ministry of Science, Education and Youth, and by the European Union-NextGenerationEU, under the research project "Breeding development of alfalfa and red clover germplasm adapted to climate changes (ALFRED BREED)" (541/23-204).

Comparative analysis of physical and morphological traits in three pear (*Pyrus* spp.) cultivars

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ABSTRACT

This study aimed to perform a comparative physical and morphological characterization of fruits from three pear cultivars, ‘Packham’, ‘Abate Fetel’ and ‘Forelle’, in order to identify traits useful for varietal differentiation and quality assessment. Fruit height, diameter (both measured using a digital caliper), fresh weight, density and skin color (measured using a colorimeter) were determined in mature fruits purchased at a supermarket. Significant differences were observed among cultivars for most parameters. Fruit height differed significantly in all cultivars, with ‘Abate Fetel’ exhibiting the greatest elongation and ‘Packham’ presenting the lowest values. Regarding diameter, ‘Abate Fetel’ fruits were significantly smaller than the other cultivars. In contrast, ‘Forelle’ showed significantly higher fresh weight and the highest ΔE values for skin color, indicating marked chromatic heterogeneity. No significant differences were detected in fruit density among cultivars. Skin color parameters clearly distinguished the three varieties, with ‘Forelle’ exhibiting pronounced chromatic heterogeneity, reflected by the highest ΔE value, in contrast to the more homogeneous appearance of ‘Packham’ and ‘Abate Fetel’. The marked differences observed in fruit height, diameter, fresh weight, and skin color demonstrate that physical and morphological parameters, particularly fruit dimensions and color heterogeneity, can be used to discriminate some pear cultivars and provide objective indicators of fruit identity and market quality. This approach represents a simple and robust tool for varietal characterization, postharvest evaluation, and quality control in pear production chains.

Keywords: colorimetry, fruit morphology, pears

Dynamics of S, Zn, and Mo uptake, transport and accumulation in *Triticum durum* Desf.

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ABSTRACT

This study evaluated the effects of three biostimulants (BlueN® - T6 and T7, Contribute ibNP – T2 and T3, and Status® - T4 and T5) combined with standard (T1, T2, T4 and T6) and reduced (T3, T5 and T7) fertilization regimes (Foskamónio 12-24-12, 150 vs. 113 kg ha⁻¹) on the dynamics of S, Zn, Mo uptake, transport, and accumulation in durum wheat (*Triticum durum* Desf.). Dry matter allocation revealed a clear shift towards aboveground organs, with T6 enhancing vegetative growth (root: 5.10 g; stem: 12.73 g), while T3 and T7 promoted greater grain biomass, indicating improved source–sink relationships. Micronutrient accumulation exhibited strong treatment-dependent responses, reflecting differential uptake and internal redistribution processes. Total S, Zn, and Mo accumulation peaked under T7 (per plant 4.06 mg, 0.092 mg, and 0.013 mg, respectively), suggesting enhanced acquisition efficiency and translocation capacity. Grain nutrient loading was particularly responsive, with maximum values per plant of 1.02 mg S, 0.029 mg Zn, and 2.25 µg Mo, indicating effective remobilization towards reproductive tissues. These patterns support coordinated regulation between biomass production and micronutrient partitioning, likely driven by treatment-induced changes in physiological sink strength and transport mechanisms. Overall, the results show that targeted treatments can simultaneously optimize yield formation and grain nutritional quality, reinforcing their potential for biofortification.

Keywords: biostimulants, mineral accumulation, mineral uptake, *Triticum durum* Desf., X-ray fluorescence

Modification of the CPVO technical protocol for DUS examination of soybean

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ABSTRACT

DUS testing (Distinctness, Uniformity and Stability) is a key procedure for the recognition and protection of new plant varieties. It enables clear differentiation from existing varieties, ensures genetic stability, and guarantees uniformity in production. Its development is closely linked to plant breeding progress and the need for intellectual property protection in agriculture. In Europe, DUS testing dates back to the 1930s, when France introduced one of the first variety control systems due to the rapid increase in wheat varieties. Regulations adopted in 1933 required varieties to be distinct and sufficiently uniform for marketing. Early assessments relied mainly on morphological traits such as plant height, ear shape, and color, evaluated through field trials. These systems formed the basis for modern standardized DUS methods, later revised in 1942 and 1960, with harmonization continuing today. Currently, the Community Plant Variety Office (CPVO) plays a central role by defining technical protocols for DUS testing in the EU. The latest soybean protocol (CPVO-TP/080/2), adopted on January 28, 2026, and in force since February 1, 2026, increases the number of characteristics to 21, adds three new traits, revises the scoring system, and updates example varieties. As in earlier versions, biochemical methods such as peroxidase analysis complement morphological traits, which remain the primary assessment basis. The reference collection is essential and includes EU-recognized varieties, those protected in UPOV member states, and other commonly known varieties. For hybrids, all components must be included to ensure reliable results.

Keywords: CPVO, DUS testing, soybean

Protein Hydrolysate–Induced Modulation of Leaf Phenolic Metabolism in Olive

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ABSTRACT

The aim of this study was to evaluate the effect of foliar application of protein hydrolysates on the phenolic profile of olive leaves ('Leccino' cultivar). The experiment was conducted in Zadar County (Preko) using a completely randomized design across 30 young trees. Foliar treatments were applied four times at 10-day intervals, starting 50 days after flowering. In addition to the untreated control, protein hydrolysates derived from pumpkin seed cake were applied, obtained by enzymatic hydrolysis using Alcalase[®] Pure + Flavourzyme[®] (H2 treatment) or Alcalase[®] Pure + Protana[™] Prime[®] (H3 treatment). The application of protein hydrolysates (PH) significantly influenced the phenolic composition of olive leaves at harvest period, with the most pronounced effects observed in the H2 treatment. Flavonoids (luteolin and apigenin) and oleacein were higher under PH treatments, particularly in H2, while glycosylated flavonoids showed a decreasing trend, with luteolin-7-O-glucoside following the pattern H2 < H3 < control. Similarly, secoiridoid precursors, including oleuropein and 11-methyloleoside, were comparable between control and H3, but lower in H2. On the other hand, oleuroside increased progressively from H2 to H3, reaching the highest values in the control. These results indicate a treatment-induced shift from precursor and glycosylated forms toward more bioactive phenolic compounds. Overall, protein hydrolysates, particularly H2, markedly modulated phenolic metabolism, potentially altering the functional properties of olive-leaf derived products.

Keywords: olive- leaves, secondary metabolism, secoiridoids, flavonoids, biostimulants

Morphological and physiological responses of *Impatiens x hybrida* plants grown under reduced irrigation regimes

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ABSTRACT

A greenhouse experiment was conducted to evaluate the effects of reduced irrigation on the morphological and physiological traits of *Impatiens x hybrida* 'SunPatiens Compact Orchid Blush', with the aim of assessing its adaptability to water-limited conditions. Plants grown in plastic pots were subjected to three irrigation levels for 45 days: 100% of field capacity (control) and two reduced levels at 80% and 60% of the control. As water availability decreased, vegetative growth was progressively suppressed, resulting in shorter and narrower plants. Although the numbers of open and wilted flowers remained unchanged, flower bud production declined significantly. Reduced irrigation also decreased the sizes of stomatal guard cells and stomatal pores, without affecting stomatal density, thus limiting the maximum total pore area available on the leaf surface. The maximum quantum yield of photosystem II (Fv/Fm) and total chlorophyll content remained stable across treatments, indicating that the integrity of the photosynthetic machinery was preserved. Total phenolic and ascorbic acid contents increased under reduced irrigation, whereas overall antioxidant activity decreased, particularly in plants receiving 60% of the control water content. Collectively, the results suggest that *Impatiens x hybrida* is sensitive to the levels of water limitation imposed in the study. However, it responds by adaptively altering its morphological and physiological traits to improve resilience and survive under both reduced irrigation regimes tested.

Keywords: antioxidant activity, phenolics, photosynthetic pigments, stomata, SunPatiens

Hvar vineyards

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ABSTRACT

Viticulture in the Republic of Croatia is a very important agricultural branch. As a labor-intensive crop, grapevines enable the employment of a larger number of people, as well as the generation of higher incomes per unit of area compared to most agricultural crops grown in Croatia. For this reason, the importance of the current intensive development of viticulture and its contribution to the economic progress of the entire country is undeniable, and the importance of viticulture is even greater if we bear in mind that it contributes to the affirmation of rural areas and the promotion of wine-growing areas. Knowledge of the actual situation when it comes to areas under vines, assortment, age of plantations, climatic and soil conditions, the potential of wine-growing geographical production areas, certain statistical data and the structure of vineyards and farms, as well as the analysis of other important aspects of viticulture is a necessary prerequisite for the further improvement and planned development of this sector. This research aims to present significant information about the Hvar vineyards and to provide a clear overview of the state and potential of viticulture on the island of Hvar.

Keywords: viticulture, vineyards, climate, wine-growing potential

Selection of the Most Efficient Pollinizers for the Cultivar Lastovka Using SSR Markers and Paternity Analysis

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ABSTRACT

Cultivar ‘Lastovka’ is an autochthonous oil-producing olive cultivar which is often planted with the dominant cultivar ‘Oblica’; however, it typically exhibits earlier flowering than most other cultivars. Despite its expanding cultivation and importance within mixed-cultivar groves, limited information is available regarding its reproductive compatibility and degree of self-incompatibility. In this study, seed paternity analysis based on simple sequence repeats (SSRs) was used to identify potential pollen donors of the cultivar ‘Lastovka’ exposed to open-pollination in a mixed olive orchard over two consecutive years. Pollination success was strongly influenced by the abundance and spatial distribution of pollen donor cultivars in the orchard, as well as by flowering overlap with the cultivar ‘Lastovka’. In the first year, ‘Cipressino’ and ‘Levantinka’ were identified as the predominant pollen donors for ‘Lastovka’. In the second year, ‘Levantinka’ remained the main pollen donor, followed by ‘Drobnica’. No embryos were identified as products of self-fertilization, indicating a strong self-incompatibility response in ‘Lastovka’. Although more distant, ‘Cipressino’ proved to be an effective pollen donor, likely due to the high number of trees producing abundant pollen. As olive is an anemophilous species, wind direction and flowering phenology play an important role in pollen transfer. The prevailing eastern and northern winds likely favored pollen flow from ‘Cipressino’ and ‘Drobnica’, while the effectiveness of pollen from ‘Levantinka’ was related to its close proximity to ‘Lastovka’. All cultivars shared several overlapping flowering days with ‘Lastovka’. Interannual differences in pollen donor contribution indicate that pollination success depends not only on genetic compatibility but also on flowering overlap, orchard design, and weather conditions, while the absence of self-fertilized embryos confirms the self-incompatibility of ‘Lastovka’.

Keywords: *Olea europaea* L., cross-compatibility, self-incompatibility, microsatellites, seed paternity assignment

Project: From grain to plant - nutritional potential of wheat and oats

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ABSTRACT

The project “From grain to plant – nutritional potential of wheat and oats (NutriWOP)” is funded by the European union-NextGenerationEU (Call for funding of institutional research projects funded from source 581 – Recovery and Resilience Facility).

During the four years of project implementation the research group of fifteen researchers from three institutions will carry out field and laboratory research in order to: (1) to examine the impact of Zn and Se biofortification on the nutritional value of wheatgrass and oatgrass juice, (2) to determine the genotypic specificity of wheat and the impact of Zn and Se biofortification on wheat resistance to storage pests, (3) to determine the morphological properties of grains of different wheat and oat genotypes, as well as the phenotypic characteristics of wheat and oat seedlings grown under controlled conditions, and (4) to select wheat and oat genotypes for further research and preparation for the commercialization of wheatgrass and oatgrass products. The innovativeness of the NutriWOP project is based on a methodology that combines agronomic (biofortification) and genetic (genotype selection) approaches to research the nutritional potential of wheatgrass and oatgrass juice. The results on amino acid and carbohydrate composition, as well as concentrations of essential minerals, will provide a holistic view of the nutritional potential of fresh juices, potentially contributing to the development of functional cereal-based food supplements. Furthermore, investigating the impact of wheat variety and Zn and Se biofortification on grain resistance to storage pests contributes to the sustainable preservation of grain quality, offering significant opportunity for the applications in the food industry.

Keywords: NutriWOP, *in-vitro* bioavailability, biofortification, Se, Zn

Do early ecophysiological responses to drought predict field performance in *Quercus suber* L. provenances?

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ABSTRACT

The increasing frequency and severity of drought events pose significant threats to Mediterranean forests, especially during early developmental stages. This study assessed whether physiological and biochemical responses to controlled drought conditions can predict the field performance of *Quercus suber* L. across four provenances representing contrasting climatic origins: France (FR3), Italy (IT13), Spain (ES11), and Morocco (MA27). Long-term field data from a provenance trial in southern Portugal were integrated with controlled drought experiments using 6-month-old seedlings. The assessment focused on traits related to leaf water status (water potential at predawn, Ψ_{pd}), gas exchange (net photosynthesis, A ; stomatal conductance, g_s ; transpiration rate, E), photochemical efficiency of photosystem II (F_v/F_m), photoprotection (total carotenoids), and cellular antioxidant enzymes activity (superoxide dismutase, SOD; ascorbate peroxidase, APX; catalase, CAT). Under moderate drought ($\Psi_{pd} \approx -2.0$ MPa), approaching a critical functional threshold, xeric provenances (ES11 and MA27) maintained higher relative water content (RWC) and F_v/F_m , the latter likely supported by stronger photoprotective and antioxidant responses. In contrast, mesic provenances (FR3 and IT13) showed early signs of functional decline, with reductions in photosynthesis and F_v/F_m , indicating the onset of non-stomatal limitations. With severe drought ($\Psi_{pd} \approx -4.0$ MPa), all provenances exhibited strong declines in A (~65%), g_s (~70%), and E (~60%), consistent with an isohydric response. Even so, xeric provenances retained comparatively higher functional integrity, while mesic provenances experienced a more pronounced impairment of C-assimilation and photochemical performance. Our findings show that physiological responses at early developmental stages (young plants) align with provenance performance observed under field conditions. This supports the use of environmental controlled experiments to assess drought resilience, highlighting C-assimilation and water-use regulation as robust indicators for the selection of climate-adapted genetic material.

Keywords: antioxidants, drought, ecophysiology, provenances, *Quercus suber* L.

Loss of volatile complexity in abnormal “starlet” flowers of *Coffea arabica* L.

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ABSTRACT

This work characterized the chemical divergences on the volatile organic compound profile (VOC) between typical and anomalous (starlet) phenotypes of *Coffea arabica* L. via GC-MS under controlled conditions. PCA indicated a category separation driven by the phenotype (PC1 = 0.384). Qualitatively, typical flowers exhibited high chemical complexity with specialized classes like terpenoids (linalool, farnesene), benzenoids (methyl salicylate, benzyl alcohol), and nitrogenous compounds (indole, methyl anthranilate). In sharp contrast, the starlet phenotype showed massive pathway repressions. While the phenotypes shared 82 compounds, typical flowers possessed 323 unique species versus only 77 in starlets, representing a 0.76 reduction in unique diversity. In addition, quantitatively, starlet-flowers underwent intense repression across chemical superclasses, ranging from a ~20-fold reduction in fatty derivatives to a >1220-fold reduction in terpenoid emissions. This collapse fundamentally altered the profile composition: whereas terpenoids dominated the typical flower signature (>0.50 of total output), fatty derivatives became the predominant group in starlet-flowers. We conclude that this phenotype represents a drastic volatilome simplification and loss of specialized attractants, likely compromising ecological signaling and impacting reproductive success in *C. arabica*.

Keywords: floral anomaly, floral scent, volatilome, non-targeted metabolomics

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Transforming Apple Bio-waste into Value-Added Products and Green Energy

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ABSTRACT

Agricultural bio-waste from fruit production represents a significant yet underutilised resource within the circular bioeconomy. Apple pomace from juice processing and pruning residues are rich in lignocellulosic fibres, fermentable sugars, and bioactive compounds, showing strong potential for conversion into value-added products such as functional ingredients, nutraceuticals, and green energy. However, these residues are still largely disposed of rather than valorised. The BioLOOP project aims to develop a sustainable, resource-efficient circular model for the valorisation of apple bio-waste using advanced technologies and data-driven optimisation. The research includes the analysis and categorisation of bio-waste streams, chemical and energetic characterisation of biomass, and the application of innovative pre-treatment methods such as high-intensity ultrasound, laser treatment, and freeze-drying. The processed biomass will be transformed into powders and extracts rich in bioactive compounds, while the remaining biomass potential will be assessed for green energy production pathways such as biofuel production and other bioenergy applications. The project will also develop practical guidelines for the sustainable utilisation of apple bio-waste within agricultural and food processing systems. The expected outcomes contribute to reducing agro-industrial waste, improving resource efficiency, and supporting the transition towards a circular and sustainable agri-food sector in Croatia and the EU.

Keywords: circular bioeconomy, apple pomace, agricultural bio-waste, bioactive compounds, biomass valorisation

Small RNA-mediated regulation of defense responses in hops against *Verticillium* wilt

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ABSTRACT

MicroRNAs (miRNAs) are short, 21–24 nucleotide-long non-coding RNA molecules that regulate gene expression by binding to target transcripts. They are involved in a wide range of biological processes, including plant defense responses to fungal pathogens. *Verticillium* wilt, caused by *Verticillium nonalfalfae*, represents a serious threat to hop (*Humulus lupulus* L.) production, leading to substantial yield losses and lacking effective control measures. The aim of our study was to investigate miRNA-mediated responses to fungal infection in hop cultivars with contrasting resistance levels. Our results revealed distinct miRNA expression profiles between resistant and susceptible cultivar, indicating a potential role of miRNAs in resistance mechanisms. In particular, elevated expression of hlu-miR160a, associated with auxin signaling, and hlu-miR319f, linked to root development, was detected in the resistant cultivar. These miRNAs may therefore contribute to defense responses against fungal infection. miRNA–target interactions during disease progression were validated using RLM-RACE. Future research will focus on functional characterization of these interactions employing CRISPR/Cas9 technology to gain deeper insight into the molecular mechanisms underlying resistance in hops.

Keywords: hops, *Verticillium* wilt, biotic stress, microRNA

Mineral composition of pear trees as affected by rootstock and tree position

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ABSTRACT

The mineral status of trees significantly influences pear fruit quality at harvest, as well as its postharvest storage potential. While mineral nutrients are fundamentally dependent on soil characteristics, they are also influenced by specific cultivar/rootstock combinations. This study aimed to evaluate the macro- and micronutrient content in the leaves of four pear cultivars ('Williams', 'Abate Fetel', 'Conference', and 'Santa Maria') grown on sloping pseudogley soil. The research was conducted in the "Agroimpex Nova" orchard in Jablanica (Gradiška, Bosnia and Herzegovina). The trees were grafted onto two different rootstocks (quince and wild pear seedling) and distributed across three plot positions (top, middle, and base) over two consecutive years. The leaf mineral content (Cu, Zn, Fe, Mn, Ca, Mg) was determined using flame atomic absorption spectrophotometry (FAAS). The concentrations of the analyzed elements ranged as follows: Cu from 0.88 mg kg⁻¹ in 'Santa Maria' to 2.17 mg kg⁻¹ in 'Abate Fetel'; Mg from 0.14 mg kg⁻¹ in 'Santa Maria' to 0.53 mg kg⁻¹ in 'Abate Fetel'; Fe from 37.74 in 'Conference' to 197.16 mg kg⁻¹ in 'Williams'; Zn from 20.90 in 'Abate Fetel' to 87.62 mg kg⁻¹ in 'Conference'; and Mn from 77.04 in 'Abate Fetel' to 522.15 mg kg⁻¹ in 'Conference'. The use of seedling rootstocks resulted in lower Ca, Mg, Mn, and Zn levels, but a higher Cu content in the leaves of the examined cultivars compared to those on quince rootstocks. The results indicated that position had a statistically significant effect on the content of all analyzed elements in 'Williams'. However, in 'Abate Fetel', position did not significantly influence Mg content; in 'Conference', Mg and Fe levels remained unaffected by position; while in 'Santa Maria', no significant effect was observed for Cu content. The results of this study define the impact of production factors (rootstock and tree position) on leaf mineral content, which in turn influences fruit quality and storage potential.

Keywords: leaf analysis, nutrient profile, seedling rootstocks, orchard

Five-year dynamics of germination and seed vigor in certified ZP maize hybrids

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ABSTRACT

Seed quality is a fundamental factor in commercial seed production, with germination capacity representing one of the most important indicators of market value. In maize seed production, maintaining appropriate storage conditions is essential to preserve both germination and seed vigor over time. According to regulations in the Republic of Serbia, certified maize seed must meet prescribed minimum germination standards, making periodic laboratory testing necessary due to the natural decline in seed viability during storage. This study examined changes in total germination and seed vigor of certified hybrid maize seed over a five-year storage period (2018–2022). Annual germination assessments were performed using the standard “between paper” method on three commercial hybrids: ZP 704, ZP 873, and ZP 600. Results are presented as average values for the analyzed seed lots. In the initial year (2018), all hybrids showed very high average total germination and vigor: ZP 704 (98.8% / 98.8%), ZP 873 (100% / 100%), and ZP 600 (98.3% / 98.3%). A moderate reduction in both parameters was recorded by 2020, with germination energy and total germination exhibiting identical values. By the final year of evaluation (2022), total germination remained above 90% in all hybrids, reaching 93.7% in ZP 704, 94% in ZP 873, and 96% in ZP 600. Although vigor values in 2022 were slightly lower than total germination in ZP 704 (92.7%) and ZP 873 (88%), hybrid ZP 600 maintained equal levels of total germination and vigor (96%). Overall, the results confirm that storage conditions complied with regulatory standards, as satisfactory germination percentages were preserved in all tested hybrids throughout the study period.

Keywords: maize, germination, vigor

High-Throughput Sequencing for the Detection of Novel Viruses in Grapevine

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ABSTRACT

High-throughput sequencing (HTS) is a highly informative tool for assessing phytosanitary status. We benchmarked the diagnostic performance of three widely used HTS analysis modalities—reference-based read mapping, *de novo* assembly with sequence annotation, and k-mer-based taxonomic classification—for the detection of known and novel grapevine viruses and viroids. Using a validated diagnostic panel of grapevines harboring diverse DNA and RNA pathogens, we generated 903 million Illumina reads and constructed 1,216 *in silico* dilution replicates spanning two orders of magnitude in sequencing depth (12.4 billion reads total). Diagnostic performance was evaluated using sensitivity and specificity across methods, databases, and sequencing depths. We discuss the relative performance of these three methods for detecting known and novel viruses. In addition, we discuss logical ensemble approaches that combine mapping, assembly, and k-mer classification for maximized diagnostic sensitivity. We present recent results on the contribution of library preparation to our result. The ensemble HTS workflow described is implemented as a virus diagnostic protocol at Foundation Plant Services, UC Davis. As a case study, we present the implementation of our protocol for composite samples of grapevine from Croatia as part of a larger collaborative investigation.

Keywords: high-throughput sequencing, grapevine viruses, detection, diversity

Effects of Salt Stress on Growth and Morphological Characteristics of Forage Pea (*Pisum sativum* var. *Arvense* L.)

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ABSTRACT

Salt stress limits the agricultural productivity of many crops, including forage pea, a legume of increasing importance in fodder production. The study was designed to investigate the impact of salt stress on the morphological features and biomass production of forage pea (*Pisum sativum* var. *arvense* L.). Four forage pea cultivars (K1 – Gold, K2 – Uran, K3 – Pablo, K4 – Pedro) were subjected to four salinity levels (S1 – control, S2 – 2.0 dS m⁻¹, S3 – 3.6 dS m⁻¹, S4 – 6.2 dS m⁻¹) and two irrigation regimes (V1 – 50% WHC, V2 – 80% WHC) in phytotron experiment during the four week vegetation period. The experiment was conducted in a randomized block design with four replications. The results showed a significant negative impact of salinity on the growth of forage pea cultivars respectively. Fresh above-ground biomass significantly decreased, root length also significantly decreased, and lateral shoot number was reduced by salt abiotic stress. Above-ground mass of forage pea significantly declined with increasing salinity levels; the biomass was the lowest under high salinity for 21.5%, under moderate salinity for 14.6%, and under low salinity for 4.5% compared to the control. The irrigation regime significantly impacted the fresh above-ground biomass of forage pea cultivars. Results indicated that cultivar had a significant effect on plant height, fresh above-ground biomass and dry above-ground biomass, and number of leaves. Cultivar Uran resulted in the highest biomass production under abiotic stress conditions. Salt stress negatively affects the growth and morphological characteristics of the investigated forage pea cultivars respectively. The combination of salinity and drought stress amplifies the negative effect of salinity. The findings demonstrate that forage pea cultivars exhibit differential responses to abiotic stress, with potentially significant implications for breeding programs.

Keywords: salt stress, drought stress, forage pea, biomass production

Descriptive Sensory Analysis of Crljenak Kaštelanski Wine

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ABSTRACT

Crljenak kaštelanski, also known as Tribidrag and Pribidrag, is considered an indigenous black grape variety from the Kaštela region. Over the last two decades, this variety has been cultivated more intensively throughout Dalmatia as a result of its revitalization. In order to characterize Crljenak kaštelanski wine, analyses were conducted using FTIR spectroscopy and sensory evaluation of the wine. The aim of the study was to determine the sensory descriptors and characteristic profile of Crljenak kaštelanski wine from the Dalmatian region, specifically from the vineyards of Zadar–Biograd, Brač, Kaštela–Trogir, and Komarna. A total of 21 wine samples of this variety were collected and analyzed from the mentioned vineyard areas. The results included the chemical composition of the wines and their sensory characteristics, which depend on the grape variety, climate, soil, and production technology. The average alcohol content of the analyzed wines was 14.87%, while the average total acidity expressed as tartaric acid was 6.49 g/L, with an average pH value of 3.61. The content of reducing sugars in most samples exceeded 4 g/L, while the average measured value was 6.69 g/L. The average contents of malic acid (0.2 g/L), lactic acid (1.26 g/L), and total volatile acidity (1.0 g/L) were the parameters that varied the least among the analyzed samples. The average Crljenak kaštelanski wine is ruby red in color, full-bodied, and characterized by fine tannins. Its aromatic profile is rich in fruity notes of dark fruits such as sour cherry, blueberry, blackcurrant, and dried plum, accompanied by aromas of wood, vanilla, and coffee, while aromas of dried red fruits and herbal notes are moderately represented.

Keywords: wine characterization, Crljenak kaštelanski, FTIR analysis, descriptive sensory analysis, Dalmatian region

Opportunities of non-conventional plant extracts in functional and technological food applications

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ABSTRACT

Deep-water pink shrimp (*Parapenaeus longirostris*) is a valuable marine resource with high economic importance for Croatian fisheries. However, it is highly perishable commodity. Post-mortem changes, combined with improper handling, can quickly lead to quality deterioration and spoilage. Therefore, ensuring adequate preservation methods is essential to maintain quality, extend shelf life and reduce economic losses associated with spoilage. The aim of this study was to evaluate the potential of plant-based extracts with pronounced antioxidant and antimicrobial properties, onion peels (*Allium cepa*), green tea (*Camellia sinensis*), rosemary (*Rosmarinus officinalis*), and oregano (*Origanum vulgare*), to improve the quality, safety and shelf life of three different shrimp products. Hydroethanolic extracts were obtained using an ultrasound bath for 1 hours at 60 °C and applied to shrimp products during different processing steps. To control melanosis, fresh shrimp were submerged in green tea extract for 20 minutes before icing and packaging. Marinated shrimp were treated with a mixture of rosemary and oregano extracts before conventional marination and packaging, while shrimp pâté was produced with onion peel extract added as a natural functional ingredient. The results showed that incorporating plant extracts significantly improved the quality of all three products. The application of green tea extract effectively reduced melanosis formation in shrimp over 11 days at 2 °C. Compared to the control (without extracts), marinated products had improved microbiological stability and slower quality deterioration. Rosemary and oregano extracts inhibited microbial growth, reduced lipid oxidation, and delayed sensory deterioration, extending shelf life by approximately 46% during refrigerated storage (4–6 °C). The addition of onion peel extract (1% w/w) in shrimp pâté resulted in lower lipid oxidation and volatile amine content while maintaining acceptable sensory properties, although no significant effect on microbial load was observed. These findings highlight the strong potential of agro-food by-products as natural and sustainable alternatives to synthetic additives in seafood preservation. Their application supports the development of clean label products, contributes to waste valorization and aligns with circular economy principles. The tested plant materials represent sustainable sources of bioactive compounds, making them suitable as natural preservatives in foods.

Keywords: by-products, shrimp, onion peels, Allium, green tea, quality, shelf life

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Soybean seed priming with Si nanoparticles to enhance resilience to drought stress

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ABSTRACT

The use of nanoparticles as a seed treatment before sowing is an effective and environmentally friendly way to increase soybean resistance to abiotic stress in the early stages of growth. The aim of this study was to examine the effect of different soybean genotypes' seed priming with nano-Si to alleviate drought stress. Seeds of five soybean genotypes were primed with solutions: distilled water (hydropriming), 500 mg L⁻¹ and 1000 mg L⁻¹ SiO₂ nanoparticles. Non-primed soybean seeds were taken as a control. Polyethylene glycol (PEG 6000) was used to induce drought stress at 0% (control), 5% (mild stress), and 10% (severe stress). A germination test was done with rolled filter paper in an incubation chamber at 25°C. The germination energy was determined on the 5th day, and total germination rate and seedling morphological parameters on the 8th day. Nanoprimering showed a significant ($p \leq 0.05$) and positive effect on the growth of soybean sprouts. The treatment with Si 1000 mg L⁻¹ gave better results than the treatment with Si 500 mg L⁻¹ for seedling fresh weight (0.74 and 0.69 g, respectively), total seedling length (12.0 and 10.0 cm, respectively), root length (6.9 and 6.1 cm, respectively), and especially shoot length (5.1 and 3.8 cm, respectively). Compared with the control, Si 1000 mg L⁻¹ increased fresh weight and the length of the above-ground part of the soybean sprouts, whereas in contrast, the lower silicon concentration (Si 500 mg L⁻¹) did not show the same positive effect. The interaction between PEG level and seed priming showed that increasing drought stress reduced soybean sprouts' growth, but priming treatments partly reduced these negative effects. Among the treatments, hydropriming (12.2 cm) and Si 1000 (12.0 cm) gave better seedling length than the control (9.4 cm). The results of the present study suggest that seed priming, especially with 1000 mg L⁻¹ Si, can help seedlings better tolerate drought stress.

Keywords: *Glycine max* (L.) Merr., priming treatment, silicon nanoparticles, drought tolerance, stress response

The potential of using sheep wool pellets in cabbage cultivation

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ABSTRACT

Approximately 553,000 sheep are raised in Croatia, and although around 1,300 tons of wool are produced annually, it is often disposed of in nature, creating an environmental problem. However, sheep wool is biodegradable, rich in nutrients, and can be used in agriculture. As a solution, the production of sheep wool pellets (SWP) has been developed, which behave similarly to organic fertilizers. A one-year fertilization experiment on cabbage was conducted, including NPK fertilizer and goat manure as comparative treatments. The aim of the study was to investigate the effect of SWP on soil chemical parameters, soil CO₂ emissions, as well as morphometric parameters and cabbage yield. Soil and plant material analyses were carried out, and the results showed no significant differences in soil CO₂ emissions. Manure and SWP reduced the decline in soil pH compared to NPK fertilizer. The observed morphometric parameters indicated that, in the case of SWP application, head height, head diameter, and yield were the lowest, while manure application resulted in the highest values of the studied parameters. Although these results are based on a one-year study, they provide insight into the potential use of SWP in cabbage cultivation; however, long-term effects on soil and the cultivation of different crops should be further investigated.

Keywords: sheep wool, cabbage, sustainability, fertilization, sheep wool pellets

Evaluation of salinity and drought tolerance in cultivated and wild olive genotypes

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ABSTRACT

Olive genotypes differed in their responses to drought and salinity, and several indicators highlighted adaptation strategies between cultivated and wild genotypes. A Mediterranean-type stress scenario was simulated by applying 150 mM NaCl and 300 mM mannitol for 21 days to three reference cultivars ('Leccino', 'Oblica', 'Koroneiki') and seven wild genotypes, evaluating morphometric, ionic and biochemical traits. Drought caused a stronger reduction of shoot length and leaf area than salinity in most genotypes, confirming shoot growth, shoot dry mass, and leaf area as primary morphological markers of water deficit in wild olives. Under salinity, the most sensitive genotypes maintained relatively low foliar Na⁺ and Cl⁻ concentrations but required a longer period to stabilize ion homeostasis, whereas more resistant wild genotypes showed faster accumulation and stabilization of Na⁺ and Cl⁻ in roots and leaves, accompanied by a moderate decline in biomass. Across all treatments, root K⁺ depletion indicated a general stress response, while maintenance or slight increase of Ca²⁺ content in leaves and roots emerged as the most energy-efficient ionic adjustment, particularly in wild genotypes. Biochemically, drought generally induced stronger superoxide dismutase activity than salinity, and SOD proved more responsive and reliable as an oxidative stress marker than proline or malondialdehyde, which showed only minor or genotype-specific changes. Notably, guaiacol peroxidase activity was induced in wild 'Piculja', PLJ 7, LA 13 and LN 11, whereas it was inhibited in most other genotypes, including all three cultivars, suggesting a distinct antioxidant strategy in selected wild olives.

Two wild genotypes, characterized by stable shoot growth, rapid ion stabilization, sustained Ca²⁺ uptake and pronounced SOD activation under both stressors, stand out as promising candidates for breeding and rootstock use aimed at improving olive resistance to combined salinity and drought.

Keywords: *Olea europaea* L., ion content, biochemical changes, abiotic stress, wild olive tree

The impact of biostimulants on antioxidant activity and potential pathogen resistance in Sour Cherry (*Prunus cerasus* L.)

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ABSTRACT

Sour cherry (*Prunus cerasus* L.) is an economically important fruit species sensitive to numerous pathogens, while plant resistance largely depends on the content of antioxidant compounds such as polyphenols and anthocyanins. The application of biostimulants represents a potential measure for strengthening the natural defense mechanisms of plants. The objective of this study was to determine the effect of a biostimulant on antioxidant activity and the content of bioactive compounds in sour cherry fruits, as well as to assess their contribution to resistance against key diseases and pathogens of sour cherry. The research was conducted during 2025 in the orchard of Fruit Growing Department at the Agricultural Institute Osijek on the varieties 'Erdi Bőtermő', 'Debreceni Bőtermő', and 'Újfehértói fürtös', including biostimulant treatment (Stim Pure AA, 2 L ha⁻¹) and untreated control. Total polyphenols, DPPH antioxidant activity, and anthocyanin content were analyzed and no disease symptoms were observed during visual assessment. The results of two-factor analysis of variance showed that the biostimulant treatment had no statistically significant effect, while the variety significantly influenced DPPH activity, inhibition and anthocyanin content. A significant variety x treatment interaction was found for DPPH values, indicating a genotype dependent response to biostimulant application. The LSD test confirmed a positive effect of the biostimulant on polyphenol content in the variety *Debreceni Bőtermő* whereas this effect was not pronounced in the other varieties. The obtained results indicate that genotype plays a key role in the formation of the antioxidant potential of the fruits, while the effect of biostimulants is limited and variable. Further research is needed to confirm their effect under conditions of higher pathogen pressure.

Keywords: sour cherry, antioxidant compounds, biostimulants, variety, pathogen

Response of Lettuce to inoculation with novel native *Trichoderma* sp. strain STP8

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ABSTRACT

Lettuce is a widely cultivated leafy vegetable with high economic importance. Increasing population growth and changing dietary habits have raised demand for higher quantities and quality produce. Sustainable production approaches include the use of beneficial *Trichoderma* fungi, as a dominant plant biostimulants and biocontrol agents, using their enzymes and certain volatile and non-volatile metabolites which act as biosynthesizers. Also, in nutrient-poor soils, they improve nutrient solubilization. Thus, seed treatment and soil incorporation near seedlings are the most common and effective methods of applying *Trichoderma*. The aim of this study was to evaluate the effectiveness of the native *Trichoderma* sp. strain STP8 (formerly *T. koningiopsis* agg. STP8) in promoting seedling growth via seed treatment and lettuce productivity in the field via soil treatment. A spore suspension (4×10^6 spores mL⁻¹) was applied. Seeds of Batavia type lettuce ‘Bataille’ was sown on April 24. Harvest took place on July 7, 43 days after planting. Biomass, marketable rosette weight and yield were measured, along with dry matter, micronutrients, and total chlorophyll. The highest rosette weight (567 and 563 g) and yield (6.30 and 6.25 kg m⁻²) were achieved with two (SP + 26 DAP) and three applications (sowing + SP + 26 DAP). The highest dry matter (6.60%) was recorded in sowing + SP treatment. Zn and Cu were highest in 26 DAP and untreated plants, while Fe and Mn increased with SP and 26 DAP treatments. Chlorophyll content ranged from 0.28 to 0.53 mg g⁻¹, with the highest value achieved with 26 DAP treatment and the lowest under triple application. Lettuce responded positively to STP8 treatments across growth stages, resulting in enhanced biomass accumulation and marketable yield. Given the variability in micronutrient composition and chlorophyll content, further research should focus on optimizing application timing and regimes to improve nutritional and antioxidative properties.

Keywords: chlorophyll content, biostimulant, green technology, *Lactuca sativa* L., micronutrients

Leaf micromorphological and metabolic adaptations to drought stress in Lamiaceae

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ABSTRACT

Progressive drought stress (70%, 50%, and 30% soil water capacity [SWC]) was investigated in four species of the Lamiaceae family: *Nepeta cataria* L., *Lavandula angustifolia* Mill., *Ocimum tenuiflorum* L., and *Perilla frutescens* (L.) Britton. The study focused on leaf micromorphological traits, including the density and size of glandular trichomes, stomatal density and size, and the thickness of the lamina, mesophyll, epidermis, cuticle, and parenchymal tissues. Essential oil (EO) and total flavonoid content (TFC) were also determined. *O. tenuiflorum*, *P. frutescens*, and *N. cataria* showed high drought sensitivity, experiencing significant reductions in biomass and leaf area (up to 54.3% in *N. cataria* at 30% SWC) and exhibiting xeromorphic adaptations such as reduced epidermis and mesophyll thickness, increased stomatal and glandular trichome density, and reduced stomatal size. Conversely, the drought-tolerant *L. angustifolia* adapted through an extreme 170.3% thickening of the adaxial cuticle to prevent water loss. Metabolic responses were highly species-specific, though prolonged severe drought generally impaired both growth and secondary metabolite production. A strong biosynthetic trade-off between EO and TFC was observed in *N. cataria* ($r=-0.95$), whereas *L. angustifolia* exhibited synergistic co-accumulation under moderate stress ($r=0.93$). In *O. tenuiflorum*, TFC was closely associated with palisade parenchyma density ($r=0.98$). Ultimately, maintaining a minimum of 50% SWC is crucial to optimize EO and flavonoid yields while preventing structural collapse.

Keywords: Lamiaceae, micromorphology, secondary metabolism, phenotypic plasticity, water deficit

Dose-dependent effects of industrial poppy-head by-product as soil amendment on *Brassica napus* growth

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ABSTRACT

The objective of this study was to evaluate the dose-dependent effects of an extruded poppy-head by-product, derived from industrial alkaloid processing, on the early growth and root system development of *Brassica napus* under controlled conditions (16 h/8 h light/dark cycle, 25/18 °C day/night). A two-week pot experiment was conducted using clay soil with five treatments: control, synthetic soil conditioner (Sedipur, 2 g kg⁻¹ soil), and three application rates of the by-product (10%, 20%, and 30% w w⁻¹) in a completely randomized design with four replicates. Soil moisture was maintained by gravimetric adjustment. Total root length, root and shoot dry weight, and root diameter distribution were measured. Treatments significantly affected root growth. Total root length ranged from 67.17 to 128.10 cm plant⁻¹, with the highest value at 20%, exceeding the control (114.57 cm plant⁻¹). Root dry weight followed a similar pattern, reaching 0.0550 g plant⁻¹ at 20% compared to 0.0405 g plant⁻¹ in the control, while the 30% treatment reduced both parameters. Fine root development (<0.2 mm) was highly sensitive, ranging from 34.72 to 82.03 cm plant⁻¹, showing stimulation at moderate doses and inhibition at higher rates. Significant treatment effects were found for root length ($p < 0.001$), root dry weight ($p < 0.05$), and fine root development, with the strongest effect in the <0.2 mm fraction ($p < 0.001$), whereas shoot dry weight was not significantly affected. Regression analysis indicated a quadratic dose-response relationship. In conclusion, the extruded poppy-head by-product significantly alters root system architecture in a dose-dependent manner. Moderate application rates improve root development and biomass accumulation, while higher doses inhibit growth. These findings highlight the importance of optimal dose selection when applying industrial organic by-products as soil amendments.

Keywords: biomass allocation, fine root development, organic waste utilization, root diameter distribution, quadratic response

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Influence of Extraction Method and Polymer Composition on Release Behavior of Protein-Loaded Alginate Microparticles

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ABSTRACT

Designing alginate-based carriers with predictable protein release requires precise control of both extract properties and polymer architecture. The objective of this study was to evaluate how extraction intensity and polymer composition influence encapsulation efficiency, swelling behavior, and protein release from calcium-crosslinked alginate microparticles. Protein-rich extracts obtained from beetroot and broad bean leaves by heat treatment at 40 °C or ultrasound-assisted extraction at 100% amplitude for 9 min were encapsulated using alginate (ALG/Ca) and alginate–carboxymethylcellulose composites (ALG/CMC/Ca) prepared by ionotropic gelation. Encapsulation efficiencies ranged from 50.53% to 64.03% for ALG/Ca and from 54.24% to 63.12% for ALG/CMC/Ca, while loading capacities were between 15.23 mL g⁻¹ and 34.16 mL g⁻¹. Polymer composition significantly affected hydration, with swelling degrees increasing from 53.30%–57.85% in ALG/Ca to 104.30%–123.80% in ALG/CMC/Ca systems. All samples showed a biphasic release profile with rapid initial release during 60–120 min followed by sustained protein release up to 1440 min. Extracts obtained by ultrasound accelerated release, whereas incorporation of carboxymethylcellulose reduced diffusion and prolonged retention. Kinetic analysis using Korsmeyer–Peppas and modified power-law models showed strong agreement with the experimental data ($R^2 = 0.97–0.99$) and indicated different release mechanisms, including Fickian diffusion ($n = 0.13–0.20$), anomalous transport ($n \approx 0.47$), and swelling-controlled release ($n \approx 1.02$). The results demonstrate that extraction conditions and polymer blending can be used as complementary design parameters for tailoring protein release from alginate-based microparticles in controlled delivery systems.

Keywords: extraction, beetroot leaves, broad bean leaves, encapsulation, microparticles

Distribution and rhizosphere potential of wild *Fragaria* spp. in Croatia for PGPR application

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ABSTRACT

Wild strawberry species (*Fragaria* spp.) are valuable genetic resources for sustainable agriculture due to their adaptability, nutritional value and tolerance to environmental stresses, particularly drought. In Croatia, three wild species occur (*F. moschata*, *F. viridis* and *F. vesca*), with *F. vesca* being the most widespread. This study, conducted within the BioJagoda – BioStrawberry project (University of Zagreb, Faculty of Agriculture, 2025–2028), aimed to determine the geographical distribution of wild *Fragaria* species and evaluate the potential of their rhizosphere microbiota as a source of plant growth-promoting rhizobacteria (PGPR), together with the planned application of characterized PGPR to support sustainable strawberry production under abiotic stress. Field surveys during the 2025 and 2026 growing season included GPS mapping and sampling of plants and rhizosphere soils from coastal (Pula), mountain (Klana) and continental (Zagreb) regions. Results confirmed the broad ecological distribution and high adaptability of *F. vesca* across contrasting climatic conditions, indicating strong phenotypic stability. Collected samples were deposited in the ZAGR herbarium and successfully maintained under controlled growth chamber conditions. Preliminary microbiological analyses revealed diverse rhizosphere bacterial communities with significant PGPR potential. Ongoing habitat suitability modelling using the MaxEnt approach integrates bioclimatic variables to predict potential distribution and identify areas suitable for conservation and cultivation. Overall, wild *Fragaria* populations represent important reservoirs of adapted plant material and beneficial microbiota, supporting their relevance for improving plant resilience, sustainable cultivation and the development of PGPR-based biotechnological applications under changing environmental conditions.

Keywords: *Fragaria vesca*, geographical distribution, rhizosphere microbiota, plant growth-promoting rhizobacteria (PGPR), sustainable strawberry production

Etiology of branch dieback of almond in Serbia

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ABSTRACT

Almonds (*Prunus dulcis* (Mill.) D.A. Webb) are one of the most popular nuts, with a high fat content that does not contribute to cholesterol formation. Although almonds are not usually cultivated in Serbia, recently several orchards have been established under suitable agroclimatic conditions. In 2024, almond growers near Belgrade reported the branches dieback due to a disease that has not yet been studied in Serbia. Eight single-spore isolates with a uniform appearance on PDA were obtained from plants with symptoms such as light spots with a dark halo on the leaves, reduced growth and internal wood necrosis. The isolates formed light grey colonies on PDA, which turned dark grey to black after 5 days post inoculation (dpi). After incubation of 20 days at 23 °C and 12 hours of light/darkness on pine needle agar, blackish, solitary pycnidia with ovoid, brown conidia (average size 29 x 12.3 µm) were visible. The morphological features resembled those of *Diplodia seriata*, which was further confirmed by ITS rDNA sequencing. A BLAST analysis of the ITS rDNA sequence of isolate 162-24 (Acc. No. PV216485) revealed 100 % identity with over 100 *Diplodia seriata* isolates from different host plants worldwide. The selected isolate (162-24) fulfilled Koch's postulates as wound-inoculated healthy branches developed black necrotic zones 15 dpi, followed by successful reisolations. The control plants remained asymptomatic. This is the first report of almond dieback in Serbia caused by *Diplodia seriata*, which is of great importance for establishing efficient disease control. This paper is the result of projects 451-03-34/2026-03/200116 funded by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia.

Keywords: almond, *Diplodia*, phytopathogenic fungi, wood necrosis, pathogenicity

Cold tolerance in maize: Linking seed germination and early photosynthetic stability under low temperature

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ABSTRACT

The aim of this study was to evaluate cold stress tolerance of six maize inbred lines at two developmental stages—seed germination and early seedling growth—in order to assess whether tolerance at the seed stage is associated with tolerance at the seedling stage, and to examine the effect of seed priming on improving germination under cold conditions. Germination was tested under optimal conditions, at 10 °C, at 10 °C with pre-soaking in water, and at 10 °C with pre-soaking in gibberellic acid solution. Chlorophyll *a* fluorescence was measured on seedlings exposed to a 24 h cold stress, with measurements taken 2 h and 24 h after the temperature drop, alongside an untreated control. Two-way ANOVA revealed significant effects of both treatment and genotype on all germination and fluorescence parameters. Cold stress reduced germination energy and final germination percentage, with the strongest reduction observed in Os622448 and Mo17, while Os7798/2 and Os6251/15 showed the highest tolerance at the seed stage. Gibberellic acid pre-treatment partially alleviated cold-induced inhibition of germination across genotypes. At the seedling stage, cold stress caused a progressive decline in the maximum quantum efficiency of photosystem II photochemistry (F_v/F_m), accompanied by increases in relative variable fluorescence at the J-step (V_j) and absorption flux per reaction center (ABS/RC), indicating closure of photosystem II reaction centers and disruption of electron transport, more pronounced after 24 h than after 2 h of stress. Pearson correlation analysis revealed a significant positive correlation between germination energy under cold stress and F_v/F_m measured 2 h after stress exposure ($r = 0.847$, $p = 0.033$), while this relationship was not maintained after prolonged stress. Principal component analysis further separated genotypes along a cold tolerance gradient consistent across both developmental stages. These results suggest that the association between seed and seedling responses is strongest immediately after stress exposure and weakens with prolonged stress, indicating that early-stage physiological traits may serve as time-dependent indicators of cold tolerance in maize.

Keywords: maize, cold stress, seed priming, germination, chlorophyll *a* fluorescence

Flowering period of hazelnut cultivars in agro-ecological conditions of Donja Zelina

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ABSTRACT

Hazelnut (*Corylus* spp.) is a fruit species showing the highest growth in terms of the quantity of declared planting material and the increase in orchard area. With 6.933 ha of recorded production areas (APPRRR, ARKOD 2025.), hazelnut represents one of the most important fruit species in terms of cultivation in the Republic of Croatia. The most represented is the autochthonous cultivar 'Istarski duguljasti' with a share of 49 %, followed by the cultivar 'Rimski' with 45 %, while other cultivars account for about 6 %, with a trend of increasing representation of newer cultivars. In order to improve pollination and address the need for introducing new cultivars into commercial orchards, a ten-year study of flowering of 11 hazelnut cultivars was conducted in the experimental orchard of the Croatian Agency for Agriculture and Food in Donja Zelina. During the research period, significant differences in the onset and duration of flowering among all observed cultivars were recorded, indicating a strong influence of climatic conditions, particularly air temperature. Analysis of the overlap in the average flowering period showed that the cultivars 'Haleški' (male inflorescence flowering duration: 35 days), 'Barcelona' (40 days), and 'Tonda Romana' (30 days) have relatively long flowering periods that overlap well with the flowering of female inflorescences of the cultivars 'Istarski duguljasti' and 'Rimski', making them potentially good pollinizers. However, at the genetic level, the cultivars 'Istarski duguljasti', 'Rimski', and 'Tonda Romana' share the S10 allele, which makes their inter-cultivar pollination suboptimal due to the occurrence of sporophytic incompatibility. Therefore, to ensure successful fertilization and more stable yields, it is recommended to include cultivars with different S-alleles in orchards, such as 'Haleški', 'Barcelona', 'Tonda di Giffoni', and 'Tonda gentile delle Langhe', whose flowering periods overlap with the target cultivars and enable effective cross-pollination.

Keywords: hazelnut, flowering period, climatic conditions, pollination

Nutritional quality of wheat seedlings biofortified with iodine

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ABSTRACT

Iodine is an essential element involved in the normal functioning of the body, particularly the thyroid gland, where it is required for the synthesis of thyroxine and triiodothyronine. The problem of insufficient iodine intake can be addressed through biofortification of crops or dietary supplements. Wheatgrass (*Triticum aestivum* L.) is rich in vitamins, minerals, and phenolic compounds and has many scientifically proven positive effects on the human body. The aim of this study was to enrich wheatgrass, which is often used as a dietary supplement due to its nutritional value, with different concentrations of potassium iodide, 40, 80 and 160 μM . Nutritional value was evaluated by measuring the amounts of vitamin C, total phenols, chlorophyll, carotenoids, total sugars, cellulose, and total antioxidant activity. Increasing the concentration of potassium iodide, used for wheatgrass biofortification, also increased the amounts of vitamin C, protein, chlorophyll, and total sugars in wheatgrass. However, high concentrations proved to be toxic as there was a decrease in wheat biomass and germination. Therefore, wheatgrass enriched with iodine due to its nutritional value has the potential for use in the daily diet or for the treatment of malnutrition.

Keywords: iodine, wheatgrass, biofortification, nutritional value

Impact of varying sowing dates on maize cob characteristics and yield

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ABSTRACT

The aim of this study was to examine the effect of different sowing dates on cob parameters and grain yield of maize hybrids with different maturity groups. The field experiment was conducted in a drought year at the Látókép Experimental Site of the University of Debrecen, using three hybrids with different FAO numbers (FAO 380, FAO 420, FAO 490) and three sowing dates (early – SD1: April 4; optimal – SD2: April 12; late – SD3: May 3). Based on the average of the hybrids, the highest values of cob weight, cob length, and cob diameter were observed at SD3, with statistically significant differences ($p < 0.05$) except for cob length. Our results showed that the highest grain weight per cob was recorded at the late sowing date (SD3) for all hybrids (FAO 380: 223.27 ± 17.21 g; FAO 420: 217.87 ± 34.57 g; FAO 490: 197.56 g; $p < 0.05$). Grain yield was also highest under SD3 (FAO 380: 11.83 ± 2.20 t ha⁻¹; FAO 420: 12.61 ± 0.27 t ha⁻¹; FAO 490: 11.79 ± 0.55 t ha⁻¹). Sowing date showed the strongest association with cob parameters in the FAO 380 hybrid (cob length: $r = 0.845^{***}$; cob weight: $r = 0.839^{***}$; grain weight: $r = 0.840^{***}$), while the strongest correlation between sowing date and yield was observed for the FAO 490 hybrid ($r = 0.837^{***}$). We concluded that later sowing (late April to early May) had a positive effect on cob length, cob weight, cob diameter, grain weight, and yield. In addition, earlier-maturing hybrids exhibited more favorable cob parameters and higher yields. Overall, optimizing sowing date plays a key role in maize yield formation. From a practical perspective, applying a later sowing date combined with shorter maturity hybrids may help achieve stable and high yield levels.

Keywords: cob parameter, hybrid, maize, sowing date, yield

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Development of innovative bio-packaging solutions by valorizing agri-food residues

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ABSTRACT

Conventional plastic packaging contributes to environmental pollution and has limitations in preserving food quality, often resulting in spoilage and waste. These challenges highlight the need for natural, bio-based alternatives. Agri-food residues (AFRs), typically treated as waste, represent a valuable source of biofibers and bioactive compounds that can be used for the development of novel packaging materials. This study focuses on the development of innovative active films from AFRs, aimed to extend shelf life while promoting a circular and sustainable approach.

Hence, various AFRs, including orange peel (OP) and selected edible flowers, were processed using an advanced wet friction milling (AWFM) technique, conducted for up to five passes, using water as a green solvent. The extracts were analyzed for total phenolic and flavonoid content, ferric-reducing antioxidant power, DPPH radical scavenging assays and total proteins to characterize their bioactivity and phytochemicals.

Owing to their high content of fibers and bioactives, the treated AFRs were subsequently used as additives for biofilm production. The results showed that AWFM is a mild and effective extraction method that enhances the release of bioactive compounds, with increasing efficiency observed as the number of passes increased. This improvement is primarily attributed to the progressive reduction in particle size, as confirmed by particle size distribution analysis and bioactivity measurements.

The obtained suspensions show strong potential for incorporation into biodegradable polymer matrices to produce active films. Preliminary tests indicated that multilayer film composed of polylactic acid and OP layers, showed better performance compared to other produced film. Further studies are needed to characterize the films.

Overall, the development of bioactive packaging solutions based on valorized AFRs represents a promising strategy for sustainable food preservation and waste valorization.

Keywords: extraction, bioactive compounds, bio-packaging, active films, solvent casting

Determination of aflatoxin content (B1, B2, G1, and G2) in different samples of commercial maize

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ABSTRACT

Aflatoxins are toxic secondary metabolites produced by fungi of the genus *Aspergillus*, which most commonly develop under conditions of elevated temperature and humidity. The most significant among them are aflatoxins B1, B2, G1, and G2, with aflatoxin B1 being considered the most toxic and a proven carcinogen capable of causing liver damage, impaired immune response, and other serious health disorders in humans and animals. Testing for aflatoxins in cereals is of utmost importance because cereals, particularly maize, are a staple food in human nutrition and are often susceptible to contamination during production, storage, and transport. This study examined the content of aflatoxins B1, B2, G1, and G2 in various samples of commercial maize to assess their safety and compliance with current regulations. The analysis included samples from populations of white and yellow Osmak, blue popcorn population, ZP Rumenka variety, yellow popcorn hybrid ZP 611K, and hybrid ZP 552B, obtained from the Maize Research Institute Zemun Polje in 2024. Determination of aflatoxin content was performed using method DML 1.29:2017, employing liquid chromatography-tandem mass spectrometry (LC-MS/MS), which provides high selectivity and reliability in mycotoxin analysis. The results showed that the concentrations of total aflatoxins in all examined samples were below 4 µg kg⁻¹, while in the yellow popcorn maize sample, the value was below 1 µg kg⁻¹. Based on the laboratory findings, it can be concluded that the analyzed commercial maize samples do not pose a risk to consumer health regarding aflatoxin content. The measured values comply with the requirements of the Regulation on Maximum Levels of Certain Contaminants in Food, published in the Official Gazette of the RS, confirming the safety and health compliance of the tested samples.

Keywords: maize, aflatoxins, LC-MS/MS, contamination, health

Acknowledgments: Investigations necessary for this study are part of the Projects grant numbers 451-03-34/2026-03/200116, 451-03-33/2026-03/200040 financed by the Ministry of Science Technological Development and Innovation of the Republic of Serbia.

Establishment of *in vitro* culture of *Castanea sativa* Mill.

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ABSTRACT

Sweet chestnut (*Castanea sativa* Mill.) is a species of considerable economic and ecological importance. Conventional propagation methods are often limited by low efficiency and phytosanitary constraints, making *in vitro* culture a valuable alternative for the production of pathogen-free plants under controlled conditions. *In vitro* culture represents a branch of plant biotechnology that includes techniques for vegetative propagation of plants on defined nutrient media under controlled environmental conditions. One of the main advantages of this approach is that plant multiplication can be initiated from a very small amount of plant material, represented by initial explants. This is particularly important for the conservation of endangered or rare species, as well as for plants that show difficulties in seed propagation.

The aim of this study was to introduce *C. sativa* into *in vitro* culture using a standard surface sterilization procedure and to evaluate the success of explant establishment. Explants were obtained from branch segments collected from the tree crown in February. The segments were placed upright in jars containing tap water and transferred to a growth chamber to stimulate the sprouting of axillary shoots. Isolated axillary bud explants were introduced into Woody Plant Medium (WPM) (Lloyd & McCown, 1980) supplemented with 0.5 mg L⁻¹ cytokinin (BAP) and 0.1 mg L⁻¹ auxin (IBA). Cultures were maintained at 21 ± 2 °C under a 16 h light / 8 h dark photoperiod. The results confirmed the feasibility of establishing aseptic cultures of *C. sativa* using a standard sterilization protocol, providing a reliable basis for further stages of micropropagation and clonal propagation of selected genotypes.

Keywords: *Castanea sativa* Mill., *in vitro* culture, surface sterilization, axillary bud, aseptic culture.

Acknowledgment: This work is supported by the Ministry of Scientific and Technological Development and Higher Education of the Republic of Srpska (19.032-968-37/25) with support from COST Action CA21157.

Yield trend in soybean VCU trials over 25 years in Croatia

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ABSTRACT

The assessment of the Value for cultivation and use of soybean varieties, for the purpose of variety registration and inclusion in the List of Varieties of the Republic of Croatia, is conducted in accordance with the Ordinance on the Registration of Varieties of Agricultural Plant (Official Gazette No. 7/2024). The testing was carried out at four experimental locations with differing climatic, pedological, and other agroecological characteristics over the period 2001–2025. The field trials were arranged in a randomized complete block design with four replications, taking into account the maturity groups of soybean varieties. According to maturity, the submitted soybean varieties are classified into groups: very early (000, 00), early (0) varieties, and medium-early (0–I, I) and late (II) varieties.

In VCU trials for the very early maturity group, 8 soybean varieties were included; in the early maturity group, 285 varieties; and 140 varieties belonged to the medium-early and late maturity groups.

The minimum annual yield in VCU trials for the very early soybean group was recorded in 2007 at 2.86 t ha⁻¹, while the maximum annual yield of 4.32 t ha⁻¹ was achieved in 2005. In the early soybean group, the minimum annual yield in VCU trials was 2.68 t ha⁻¹ in 2005, and the maximum was 4.96 t ha⁻¹ in 2010. For medium-early and late soybean varieties, the minimum annual yield in VCU trials was recorded in 2012 at 2.04 t ha⁻¹, while the maximum yield of 5.60 t ha⁻¹ was achieved in 2010.

The obtained linear regression models are not representative, as the coefficients of determination for all soybean maturity groups are very low. For the very early maturity group, the coefficient of determination is $R^2 = 0.2372$; for the early maturity group, $R^2 = 0.0002$; and for the medium-early and late maturity group, $R^2 = 0.0041$. A possible reason for this outcome is the pronounced fluctuations in yield levels across years of VCU soybean variety trials, which can be attributed to the influence of numerous environmental and production factors.

Keywords: soybean, linear trend, grain yield, variety testing

Reduction of climate change impacts by applying mulching in red pepper (*Capsicum annuum* L.) cultivation

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ABSTRACT

Pepper production is becoming increasingly challenging due to climate change and frequent high temperatures. Extreme heat negatively affects flower fertilization, fruit set, and overall fruit quality, often leading to reduced yields. Lack of rainfall and more frequent drought periods further complicate the maintenance of optimal soil moisture and increase irrigation costs. Variable weather conditions also contribute to greater plant stress and the occurrence of diseases, making pepper production more uncertain and demanding for growers. With the aim of reducing the effects of climate change, the Agriculture Extension Service “Sombor” established a field trial with different mulching treatments: control, black plastic mulch, black plastic mulch with hay, black plastic mulch with straw, soil without plastic mulch with hay, and soil without plastic mulch with straw. The trial was set up at the experimental field and the selected variety was *Slonovo uvo*. The highest total yield was recorded in the treatment with black plastic mulch combined with hay, reaching 33.0 t ha⁻¹, along with the highest yield of first-class fruits at 31.25 t ha⁻¹. This treatment also resulted in the highest number of fruits per hectare, totaling 250,000. Additionally, the black plastic mulch with hay treatment showed a significantly lower yield of second-class fruits (1.75 t ha⁻¹) compared to the standard application of black plastic mulch. The obtained results clearly indicate that the application of mulching has a significant and positive effect on pepper production. This agrotechnical practice contributes to the preservation of soil moisture, reduction of temperature stress, and improvement of conditions for plant growth and development. Furthermore, mulching can lead to higher yields and improved fruit quality. Based on the obtained results, mulching can be considered an effective measure for mitigating the negative impacts of climate change in pepper production.

Keywords: pepper, climate change, mulching, agrotechnical measures, yield

Session 2

Animal Production and Food Quality

Fermentation Characteristics of Grass Silage in Croatia

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ABSTRACT

Grass silage plays a significant role in the nutrition of ruminants in Croatia. However, there is very few or almost no systematic data on its fermentation indicators. This paper therefore presents the results of a five-year analysis (2020–2025) of grass silage samples collected from commercial farms in 15 Croatian counties and analysed using the FT-NIR method. A total of 204 samples were analysed, dry matter (DM), pH, organic acids, ammonia nitrogen (NH₃-N), fibre fractions (ADF, NDF), and fermentation indicators were determined. The average DM content was 356 g kg⁻¹ DM with an average pH value of 4.48, indicating predominantly lactic acid fermentation. Lactic acid was the dominant fermentation product (75.0 g kg⁻¹ DM), while the amount of acetic acid averaged 18.8 g kg⁻¹ DM. Butyric acid, an indicator of clostridial fermentation, averaged 3.8 g kg⁻¹ DM, with the maximum recorded in 2022. The average NH₃-N concentration of 12.1 g kg⁻¹ DM indicates moderate proteolysis. Notably, there was a marked improvement in fermentation parameters between 2020 and 2021. Dry matter decreases from 434 to 338 g kg⁻¹ and in pH from 4.92 to 4.35, indicating that the material before ensiling in 2020 was overdried, resulting in poor fermentation. From 2021 onwards, the parameters stabilised, lactic acid dominated, and the level of butyric acid remained low (< 3.2 g kg⁻¹ DM), with a temporary increase in 2022. Regionally, most samples originated from Koprivnica-Križevci County (60%), with smaller proportions from Osijek-Baranja and Bjelovar-Bilogora Counties. Therefore, it can be concluded that FT-NIR has proven to be a suitable tool for rapid and routine determination of qualitative and fermentation parameters of grass silage in large sample batches and can be recommended for wider and systematic application in practice.

Keywords: grass silage, fermentation characteristics, FT-NIR

Nutritional value and limitations of soybean storage residues in animal nutrition

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ABSTRACT

Soybean storage residues, which include broken grains, fines (dust), damaged grains, and mechanical impurities resulting from handling and storage, represent a heterogeneous raw material with variable nutritional and hygienic quality. The chemical composition of broken grains generally matches that of whole grains (36–40% crude protein, 18–20% fat), but the increased surface area exposed to air accelerates the oxidation of unsaturated fatty acids and raises the risk of mold development. Fines and dust are particularly risky due to their greater moisture-binding capacity, faster heating, and potentially higher concentrations of mycotoxins. The main safety risks are associated with mycotoxin contamination (especially from *Fusarium* and *Aspergillus* genera), lipid oxidation, and significant variability in nutritional composition between individual lots. An additional problem with raw grains is the presence of anti-nutritional substances, primarily trypsin inhibitors and increased urease activity, which require appropriate heat treatment before inclusion in pig and poultry diets. Ruminants show greater tolerance to quality variations, but mycotoxin control is also necessary for them due to the possible transfer of aflatoxin M1 to milk. The use of stored soybean residues in animal feed can be economically justified only with systematic laboratory control (moisture, crude protein, fat, mycotoxins), technological stabilization, and limited storage time. Without analytical verification and standardization of quality, such feeds do not represent a reliable source of protein but pose a potential risk to animal health and the safety of products of animal origin.

Keywords: soybean storage residues, nutritional value

Sea buckthorn (*Hippophae rhamnoides* L.): A versatile agricultural plant in the treatment of diabetic cataracts

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ABSTRACT

The aim of the study was to analyse the interaction between diabetes mellitus II. type (DM2T) and supportive treatment with sea buckthorn berries (SB) (*Hippophae rhamnoides* L.), which has historically been cultivated in the Slovak Republic as a traditional agricultural plant used in households and in folk medicine. For a long time, SB was one of Slovakia's forgotten plants. Nevertheless, modern research has confirmed its potential due to its rich content of bioactive compounds with antioxidant effects in the treatment of certain diseases. Today, it can be considered a newly discovered agricultural crop.

Zucker diabetic fatty (ZDF) rats in the age of 14 weeks used in the study, a suitable animal model for mirroring human DM2T, were divided into groups and treated once daily by gastric gavage with diabetic medication metformin (150 mg/kg, n=10), SB in the dose of 500 mg/kg (n=10) and 1000 mg/kg (n=10) directly to the stomach. Untreated control diabetic group (n=10) received only water using gastric gavage. The experimental intervention lasted 16 weeks. Opacification of lens was examined using mydriatic drops containing 2% homatropine and 0.5% tropicamide into conjunctival sac and was evaluated in five categories (A-clear lens, B-small vacuoles, C-cortex haziness, D-hazy cortex and dense nuclear opacity, E-mature cataract). In enucleated lens the sorbitol concentration by enzymatic analysis was determined. Cataract developed more frequently in the control untreated group of rats. SB and metformin treatment showed anti-cataract activity. We found decrease in sorbitol accumulation in all treated group in comparison to the untreated control. Metformin alone had the most significant effect on decrease in sorbitol accumulation in lens. Regular monitoring of blood glucose from the tail vein showed significant decrease of values in all treated groups in comparison to the untreated control. Probably, the main anti-cataractogenic mechanism consists in hypoglycaemic effect of the SB polyphenols that results in inhibition of the polyol (sorbitol) pathway. Regular consumption of SB primarily in young age could delay onset of metabolic disease, improve diabetic symptoms and are suitable co- alternative in therapy of DM2T.

Keywords: cataract, diabetes mellitus, rats, Sea buckthorn

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Sustainable Use of Olive Oil Residues in Ruminant Diets for Methane Reduction and Product Quality

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ABSTRACT

Livestock production contributes significantly to greenhouse gas emissions worldwide, particularly methane generated during enteric fermentation in ruminants. At the same time, the olive oil industry produces large amounts of residues such as olive pomace, olive cake, pits and olive mill wastewater, which are often considered environmental burdens and doesn't get used properly. However, these by-products contain very valuable compounds, including polyphenols and residual lipids, that may influence rumen processes. This study explores the potential of olive oil industry residues as feed components in ruminant diets, with a focus on their role in methane reduction. Findings from both in vitro and in vivo research suggest that their inclusion can lower methane emissions without compromising animal performance. This effect is mainly linked to the activity of polyphenols against methanogenic microorganisms and protozoa, as well as the influence of fatty acids on rumen fermentation pathways. Also, in addition to this, improvements in the fatty acid composition of milk and meat have been reported. Overall, the use of olive oil residues in animal feeding offers a practical approach to managing agro-industrial waste while supporting more sustainable livestock production.

Keywords: olive oil residues, methane emissions, ruminant nutrition, sustainable agriculture

Quality parameters of yogurt fermented from a milk-whey mixture

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ABSTRACT

The aim of this study was to evaluate the quality parameters of yogurt produced from a milk–whey mixture (75:25, v/v), with emphasis on physicochemical properties, texture, and mineral composition. Samples were produced using this formulation and analyzed in duplicate over 21 days of storage. The analysis included composition (moisture, total solids, protein, fat, carbohydrates, and ash), textural parameters obtained by texture profile analysis, and mineral content (iron (Fe), zinc (Zn), magnesium (Mg), calcium (Ca), phosphorus (P), sodium (Na), and potassium (K)). The elements were determined using inductively coupled plasma–optical emission spectroscopy (ICP-OES) following microwave digestion. The incorporation of whey resulted in a reduction of fat content (from $3.38 \pm 0.05\%$ in control yogurt to $2.34 \pm 0.19\%$ in functional yogurt), contributing to the development of a lower-fat product. However, whey addition influenced the structural properties of the yogurt, resulting in a weaker gel network and reduced firmness (90 ± 1.41 in control yogurt vs. 58 ± 5.66 in functional yogurt at day 21). Mineral analysis showed slight differences between samples; for example, calcium content was 977.6 ± 114.5 mg/kg in control yogurt and 985 ± 314 mg/kg in functional yogurt, while zinc content was 2.63 ± 0.43 mg/kg and 2.30 ± 0.72 mg/kg, respectively, with no statistically significant differences ($p > 0.05$). In conclusion, whey incorporation enables the production of a functional, reduced-fat yogurt, although it negatively affects texture. Further optimization is recommended to improve technological properties while maintaining nutritional benefits.

Keywords: milk-whey, yogurt, physicochemical properties, texture, minerals

Field pea forage quality under rainfed conditions as affected by tillage system and organic amendments

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ABSTRACT

Field pea forage quality can be influenced by tillage practices and soil amendments, although information on their effects under rainfed conditions remains limited. This study aimed to determine the effects of tillage intensity and organic amendments on the chemical composition and selected antinutritional factors of field pea forage. The split-plot field trial was established in autumn 2020 on Stagnosols under rainfed conditions in continental Croatia. The main factor was the tillage system, which included conventional tillage (ploughing) and conservation tillage (minimal soil disturbance). Each tillage system consisted of three subtreatments: control (no amendment), farmyard manure (40 t/ha), and biochar (40 t/ha). Field pea forage biomass was harvested in 2024. Reduced tillage significantly increased contents of crude protein (228.97 vs. 209.18 g/kg DM) and water-soluble crude protein (107.89 vs. 99.85 g/kg DM), but decreased starch content (244.38 vs. 275.80 g/kg DM). It also increased contents of phytic acid (0.82 vs. 0.73 g/kg DM) and fiber-bound tannins (4.71 vs. 3.85 mg/g DM). Farmyard manure increased crude protein compared with biochar and control (241.09 vs. 208.07 g/kg DM), with the highest crude protein recorded under conservation tillage with manure (259.01 g/kg DM). Moisture, ash, crude fat, sugars, detergent fiber fractions, trypsin inhibitor activity, and free, protein-bound and total tannins were not significantly affected by treatments. In conclusion, conservation tillage combined with farmyard manure improved the protein value of field pea forage, although this was accompanied by lower starch and slightly higher contents of some antinutritional factors.

Keywords: legume, environmental friendly management, crude protein, antinutritional factors

Acknowledgement: This work was supported by the PRIMA Foundation through the “Soil Health and Agriculture Resilience through an Integrated Geographical information systems of Mediterranean Drylands” project (GA number 2211).

Chemical characterization of bee bread and antimicrobial activity of its beneficial microbiota against pathogenic bacteria

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ABSTRACT

The production of bee bread (BB) is a complex biological process in which bee pollen undergoes a fermentation process primarily carried out by the lactic acid bacteria (LAB). This study aimed to chemically characterize BB samples, isolate the beneficial microbiota and investigate their enzymatic activity, antibiotic resistance and antimicrobial effect against major foodborne pathogens causing diseases in humans (*Salmonella enterica*, *Escherichia coli*, *Listeria innocua*, *Staphylococcus aureus*). Pooled BB samples were collected from three experimental colonies of Carniolan bee (*Apis mellifera carnica*) and analysed by infrared (FTIR-ATR) spectroscopy, while the isolated beneficial microbiota was analysed by molecular methods (MALDI-TOF MS, rep-PCR) and further biotechnologically (enzymatic activity, antibiotic resistance, antimicrobial activity) investigated. The results of FTIR-ATR analysis showed unique spectral profiles of analysed BB samples, and the assignment of absorption bands provided insight into the main BB constituents (water, carbohydrates/sugars, lipids and proteins). The spectral features revealed a similar chemical composition, indicating a similar botanical origin of investigated BB. The abundance of LAB in the BB samples ranged from $4.1 \times 10^4 \pm 2.0 \times 10^3$ CFU/g to $1.6 \times 10^5 \pm 9.4 \times 10^3$ CFU/g. The isolates obtained from BB were identified as *Apilactobacillus kunkeei* (fructophilic LAB). Additional rep-PCR analysis revealed significant intraspecies variability. The isolated strains (n=22) exhibited significant proteolytic enzymatic activity and intrinsic resistance to vancomycin and kanamycin. A strong inhibitory effect was observed against all tested pathogenic bacteria (*S. enterica*, *E. coli*, *L. innocua*, *S. aureus*), ranging from strong to complete inhibition, with stronger antimicrobial activity against Gram-positive bacteria.

Keywords: bee bread, FTIR-ATR chemical characterization, beneficial microbiota, antimicrobial activity, pathogenic bacteria

Acknowledgements: This research was funded by the Paying Agency for Agriculture, Fisheries and Rural Development of the Republic Croatia (APPRRR, class: 320-09/23-07/0011) within the frame of the project entitled “Chemical, molecular and antimicrobial profiling of bee pollen, bee bread and propolis (Bee-PROfiling)” (2024-2025).

Selection potential of recapping and suppressed reproduction of *Varroa destructor* in the Carniolan honey bee (*A. m. carnica*) population in Croatia

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ABSTRACT

The aim of this study was to determine the dynamics of *Varroa destructor* reproduction and the occurrence of brood recapping behaviour in the Carniolan honey bee (*Apis mellifera carnica* Poll.) population, and to evaluate their relationship and selection potential. The research was conducted from 2016 to 2022 on a total of 235 colonies included in a breeding program. Four generations were monitored under partial mating control at a mating station with 96 drone-producing colonies. In addition to traits related to *Varroa* resistance (MNR, hygienic behaviour, and recapping), the selection program also included productive and behavioural traits (honey production, swarming tendency, and defensive behaviour).

A total of 4,858 singly infested worker brood cells were analysed. The mean mite fecundity (number of viable female offspring) was 1.37, while the mean fertility (total number of female offspring) was 2.88 per cell. No significant differences ($p > 0.05$) were found in mite fecundity or fertility between recapped and non-recapped brood cells. Across the observed generations, a slight increase was recorded in the average level of suppressed mite reproduction as well as in the frequency of brood recapping.

An additional artificial uncapping experiment demonstrated that artificial opening of brood cells significantly increased the proportion of mites with reduced fecundity. It is assumed that opening of cells may stimulate mites to enter brood cells in which reproduction will not be successful. No brood cell containing offspring without the presence of the foundress mite was recorded in this study, indicating that mites which had initiated reproduction did not abandon temporarily opened cells, except in cases where the brood was completely removed by the bees. In conclusion, the results indicate that brood recapping behaviour can be increased over several generations of selection, whereas the increase in specific *Varroa* resistance expressed as MNR (mite non-reproduction) was less pronounced.

Keywords: resistance, *Varroa*, honey bee, recapping, mite reproduction

From Fresh to Frozen: Advanced Methods for Meat Freshness Evaluation

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ABSTRACT

Meat freshness plays a key role in food safety, consumer trust and market competitiveness, yet its reliable assessment remains challenging. Common approaches, such as visual inspection and expiration dates, provide only limited insight into product history. At the same time, there is growing demand for clearer information on whether meat has been previously frozen and how long it has been stored. This is particularly problematic in retail settings, where fresh and frozen–thawed products can appear visually identical, increasing the risk of mislabelling. Current methods are often non-standardized and rely on single indicators, which do not fully reflect the complexity of quality changes during storage. The SiMPSOM project addresses these limitations by integrating physicochemical, molecular and microbiological analyses into a unified framework. It investigates pork, poultry and beef during storage, both in controlled conditions and real retail environments, including domestic and imported products to reflect market reality. Quality changes are assessed using multiple complementary indicators. Physical and oxidation markers track progressive deterioration, while DNA integrity analysis provides insight into freeze–thaw history, as frozen–thawed meat shows greater fragmentation compared to fresh samples. These changes are detected using PCR and qPCR methods. At the same time, qPCR is used to monitor the presence and dynamics of specific pathogens, linking quality assessment with food safety. All data are integrated using multivariate statistical approaches and machine learning, where methods such as principal component analysis and classification models will enable reliable differentiation between fresh, refrigerated and frozen–thawed meat with high accuracy. Overall, the project proposes a more comprehensive approach to freshness evaluation, supporting more accurate labelling and improving transparency, safety and consumer confidence.

Keywords: meat, freshness, cold storage, DNA, machine learning

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Nitrogen Use Efficiency (NUE) in Dairy Cows: Proportion of Dietary Nitrogen Transferred into Milk

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ABSTRACT

Nitrogen use efficiency (NUE) in dairy cows is a key indicator of productive performance and environmental sustainability. The aim of this study was to evaluate NUE and identify the main influencing factors, including feed composition and breed structure, across dairy farms in Kosovo. The study assessed the proportion of dietary nitrogen converted into milk across 75 dairy farms in Kosovo, distributed across five regions, during 2023–2024. The selected farms each maintained more than 10 lactating cows and were managed under closed dairy production systems, including Simmental, Holstein, and Montbéliarde breeds. From each farm, three composite samples of total mixed ration (TMR) ($n = 225$) and three fresh milk samples ($n = 225$) were collected. Nitrogen content in feed and milk was determined using the Kjeldahl method, and NUE was calculated as milk nitrogen relative to total dietary nitrogen. Feed nitrogen content ranged from 1.92% to 3.67%, while milk nitrogen ranged from 0.455% to 0.846%. NUE ranged from 14.34% to 34.49%, with a mean of 24.7%, indicating that more than 75% of dietary nitrogen was not converted into milk. ANOVA showed significant differences among farms ($p < 0.05$), mainly influenced by feed composition and breed structure. Farms using balanced rations based on maize silage, grass silage, and protein concentrates achieved higher NUE (27–30%) compared to those relying on dry forages (18–22%) ($p < 0.05$). Pearson correlation analysis showed a moderate positive relationship between feed and milk nitrogen ($r = 0.40$ – 0.50 ; $r^2 = 0.16$ – 0.25) and a weak negative relationship between nitrogen intake and NUE ($r \approx -0.30$; $r^2 \approx 0.09$). In conclusion, optimizing feed composition and protein balance can significantly improve NUE in dairy systems, reducing nitrogen losses and environmental impact.

Keywords: Nitrogen Use Efficiency (NUE), dairy cows, feed types, nitrogen losses, sustainable milk production

Molecular Genetic Characterization of the Dalmatian Bušak-Type Horse

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ABSTRACT

The objective of this study was to assess the population structure of small warmblood horses of the Dalmatian Bušak type (DB) in comparison with Croatian Coldblood (HH), Lipizzan (L), and Međimurje horse (M), as well as to evaluate maternal diversity in comparison with the Bosnian Mountain Horse (BMH) using molecular genetic approaches. A total of 113 individuals were included in the population structure analysis (DB = 21, HH = 32, L = 40, M = 20), while 15 previously reported BMH haplotypes were used for comparative mtDNA analysis. Seventeen microsatellite markers were used for population structure inference, while mitochondrial DNA (mtDNA) variation was analysed to clarify maternal relationships among populations.

Population structure was analysed using STRUCTURE software, and the most informative number of clusters (K) was determined using the ΔK method of Evanno. Haplotype networks were constructed using PopART. The mtDNA analysis identified 10 haplotypes in DB horses, distributed across 8 haplogroups according to Achilli classification, with one haplotype shared with the BMH population. This indicates moderate to high maternal diversity and suggests historical gene flow and partial common ancestry.

Population structure analysis (K = 2–4) demonstrated clear genetic differentiation of L, reflecting their controlled breeding history, while HH and M exhibited admixed profiles. DB were predominantly genetically homogeneous, with only 2 of 21 individuals exhibiting admixed genetic structure, characterized by ancestry components shared mainly with HH and to a lesser extent with M.

These results indicate that DB represent a genetically diverse and partially differentiated population shaped by both local breeding practices and historical admixture. Despite the absence of strict breeding control, their genetic identity remains detectable. The findings highlight the importance of implementing targeted conservation and breeding programs to preserve the genetic variability and authenticity of this small warmblood horse population.

Keywords: Dalmatian Bušak type horses, population structure, mitochondrial DNA, genetic diversity

***In situ* gene banks – the foundation for the preservation of indigenous pig breeds in the Republic of Croatia**

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ABSTRACT

The aim of this work is to point out the possibility of preserving autochthonous pig breeds in the Republic of Croatia through the formation of gene banks *in situ*. Intensive selection to improve the reproductive and productive properties of pigs has resulted in the deterioration of their biological properties and weakening of resistance to diseases, housing conditions and muscle and fat tissue qualitative traits. In order to improve these traits, different selection strategies are being developed, including importing the alleles associated with disease resistance, adaptive traits and meat quality, which, in most cases, can be found in autochthonous pig breeds. Although autochthonous breeds make only 5% of the total pig population, almost 85% of the genes that contribute to sustainable biodiversity have been preserved in them. In order to preserve this valuable reservoir of favourable genes and alleles, it is not enough to preserve genetic material *in vitro*, as is done in most cases today (isolated DNA, frozen sperm), but it is also necessary to preserve living animals through *in situ* gene banks. This form of preservation of indigenous breeds has been introduced into the legislation of the EU and the Republic of Croatia, and the basic principle is that highly valuable breeding individuals of indigenous pig breeds are kept in conditions that are characteristic of their breeding and that, in combination with the application of molecular genetic methods, selection procedures are carried out with the aim of preserving the most valuable qualitative and quantitative characteristics. The advantages of this method of preserving indigenous breeds are reflected in the preservation of live animals and relatively quick and simple renewal of breeds, while the risks are primarily focused on biosecurity threats (African swine fever - ASF, etc.). Given the degree of threat to indigenous pig breeds in the Republic of Croatia and the implementation of breeding and selection procedures on these breeds to date, we believe that the formation of an *in situ* gene bank would give new impetus to the development and sustainability of these breeds in our conditions.

Keywords: pigs, indigenous breeds, *in situ* gene banks, biodiversity, sustainability

Impact of Animal Welfare and Chronic Stress on Beef Quality Traits

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ABSTRACT

The GeSt Beef project* aims to improve understanding of the relationships between animal welfare, chronic stress, and beef quality through a multidisciplinary approach that includes on-farm monitoring, transport and pre-slaughter assessments, physiological and behavioural measurements, and detailed meat quality evaluation. In this context, a key objective is to assess the impact of animal welfare conditions and chronic stress on beef quality, with particular emphasis on physiological stress indicators and their relationship with technological and sensory properties of meat. The study will integrate measurements of stress biomarkers (e.g., cortisol levels in blood, saliva, and hair), behavioural observations, and carcass/meat quality traits including pH, colour, water-holding capacity, and tenderness. The expected outcomes include identification of associations between chronic stress indicators and variability in meat quality traits, as well as improved understanding of how pre-slaughter conditions influence technological and sensory properties of beef. The project is expected to contribute to the development of objective indicators for welfare assessment and to provide a scientific basis for recommendations aimed at improving animal welfare and ensuring consistent beef quality within sustainable production systems.

Keywords: animal welfare, chronic stress, beef quality, cortisol, meat traits

Acknowledgment: This work has been fully supported by the Croatian Science Foundation under the project GeST Beef UIP-2025-02-8811

Effect of Feeding Selenium-Biofortified Maize and Soybean on Production Performance and Hematological Profile of Sheep

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ABSTRACT

Selenium is an essential trace element involved in antioxidant defense, and its deficiency leads to metabolic disorders, reduced productivity, health issues, and economic losses of sheep. In many European regions, including continental Croatia, low soil selenium levels result in inadequate dietary supply in sheep - lamb production systems. Its role is especially critical during late gestation and early lactation, periods characterized by markedly increased metabolic demands. The aim of this study was to evaluate the effects of different forms and levels of dietary selenium on production performance and hematological parameters in sheep during late gestation and lactation. Thirty late gestation Merinolandschaf sheep were included in a five-month study conducted on a commercial farm in Eastern Croatia. Sheep in late gestation were allocated to three groups, and after lambing, lambs remained with their mothers throughout the suckling period until the lambs reached slaughter weight. Animals were housed in separate pens with *ad libitum* access to water. Feeding was based on hay and concentrate mixtures. The control group (I) received a standard diet supplemented with inorganic selenium (0.319 mg/kg DM), while the experimental groups (II and III) were fed diets containing selenium-biofortified corn and soybean in form organic selenium, providing either equivalent or higher selenium levels compared to the control (0.364;0.506 mg/kg DM). The results of the study showed that the form and level of selenium had no significant ($P > 0.01$) effect on the evaluated parameters. Body weight and morphometric traits were not affected by treatment, whereas time had a significant effect on their changes during late gestation and lactation ($P < 0.01$). A significant group \times time interaction indicates treatment-related differences in temporal dynamics, with group II (body weight) and groups II and III (morphometric traits) showing more favorable trends over time, but with no statistical significance. Hematological and differential leukocyte parameters were also largely unaffected by treatment, with exceptions of *mean corpuscular volume* (MCV), while time significantly influenced most variables ($P < 0.01$). Observed group \times time interactions for selected parameters suggest differences in physiological and immune responses over time, with a general tendency toward more favorable patterns in groups II and III.

Keywords: selenium, sheep, late gestation, lactation, hematological parameters

Acknowledgment: Note: This work was funded by the Croatian Science Foundation through the project HRZZ-IP-2022-10-3803.

Heat stress resilience in Holstein cows: milk yield response to THI

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ABSTRACT

Heat stress (HS), commonly quantified using the temperature-humidity index (THI), represents a critical challenge in intensive dairy production systems, impairing physiological homeostasis and animal productivity. The aim of the study was to analyze the impact of THI on daily milk yield with a particular focus on the temporal dynamics and delayed responses following acute exposure. The investigation was conducted on 169 dairy cows (Holstein) across parities 2 to 6, comprising a total of 28,285 daily milk records (average milk yield: 38.72 kg). Environmental conditions (temperature, humidity) were continuously monitored using data loggers installed across three locations on a single farm (barn, outdoor passage, and milking parlor), revealing an average THI of 70 and maximum values reaching 81.5. Linear models indicated a weak negative, but statistically significant correlation between THI and milk yield ($r = -0.061$; $p < 0.001$), corresponding to an estimated decrease of 0.13 kg per unit increase in THI. After including days in milk and parity in the multivariable model, the effect of THI was no longer significant, suggesting that strong physiological factors influence daily milk yields. Consequently, the dynamics of HS were evaluated using an event-based approach, defining acute exposure if $\text{THI} \geq 78$. Results demonstrated the absence of an immediate response on the first day of exposure, followed by a significant delayed decline: Day +2 (-0.45 kg), Day +3 (-0.40 kg), with the peak reduction occurring at Day +4 (-1.12 kg), and a persistent effect through Day +5 (-0.74 kg). Substantial variability in individual cow responses, potentially indicates to a genetic component underlying heat stress resilience, providing a basis for future analyses. In conclusion, the impact of heat stress on milk yield is characterized by a distinct temporal lag that is not captured by simple linear models. These findings emphasize the necessity of integrating delayed response into analysis of daily milk records.

Keywords: temperature-humidity index, milk yield, heat stress resilience

Acknowledgement: This research was funded by European Union - NextGenerationEU within the project “NEXT GENERATION ANIMAL PRODUCTION” (grant number NPOO.C3.2.R3- I1.04.0141)

Factors influencing the fatty acid profile of Holstein-Friesian cattle

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ABSTRACT

Milk fatty acids (FA) determine quality of dairy product, and they are influenced by multiple factors. This study aimed to evaluate the effects of lactation stage (LS), parity (PAR), milk yield (MY), total bacterial count (B), and somatic cell count (SC) on the milk FA profile of 173 Holstein–Friesian cows. Samples were collected from one commercial dairy farm, where cows were housed on a concrete floor, fed a total mixed ration (TMR), and milked three times daily. Each sample represented a different individual cow, with no repeated measurements included, ensuring independence of observations. A total of 25 FA was determined using gas chromatography. General linear model was used, and statistical significance was set at $p \leq 0.05$. The results indicated that MY showed the strongest effect on the majority of analysed FAs, including two the most abundant FAs: C16:0 ($p \leq 0.01$) with 38.1% and C18:1n9 ($p \leq 0.001$) with 23.63% of total milk FA. Notably, MY was the only factor that showed a significant effect ($p \leq 0.05$) on C18:2n6-trans (linoleic acid) encompassing linoleic acid and its various isomers, including conjugated linoleic acid (CLA), which is known for its health benefits. LS and PAR significantly affected short-chain FAs and certain monounsaturated fatty acids (MUFA). The B had a significant effect on long-chain FAs, whereas no significant influence of SC was observed. The coefficient of determination (R^2) indicated a moderate level of explained variability (0.04-0.25). This can be attributed to the biological nature of FA synthesis: short- and medium-chain FAs are synthesized *de novo* in the mammary gland, whereas long-chain FAs are largely derived from circulating lipids. Consequently, relatively low R^2 values are expected due to high biological variability, individual differences, and the influence of additional factors such as diet, genetics, season, and metabolic status. In conclusion, milk FA composition in this study was primarily influenced by MY and LS, while PAR and B exhibited more limited effects.

Keywords: milk quality, variability, fatty acids, Holstein-Friesian cows

Comparison of Pedigree-Based and Genomic Inbreeding (F_{ROH}) in Holstein Cattle Using Pedigree and SNP Data

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ABSTRACT

The two most commonly used methods of estimating inbreeding and structural relationships in livestock populations include pedigree and genomics. However, there can be significant differences between these types of estimations based upon the quality and quantity of the available data as well as the methodologies employed. This research aimed to evaluate the pedigree structure in a Holstein population from a Croatian dairy farm and to compare pedigree-based to genomic estimates of inbreeding. Production and pedigree database records were collected by the national production and pedigree databases. These included a total of 576,251 production records and 1,201,074 pedigree records. Following record filtering, 6,337 cows were selected and utilized for subsequent analysis. Each cow had a total of 14,732 records, and each had a pedigree constructed through five generations. Thus, a total of 21,779 animals formed the basis of the pedigree. A marked improvement was observed in pedigree completeness as generations progressed, providing greater confidence in genealogy within later generations. Analysis of pedigrees found increased inbreeding over time, with a maximum average inbreeding coefficient of 0.0142. The approximate rate of inbreeding was calculated to be about 0.002 (ΔF). Additionally, the effective population size (N_e) of the population was calculated to be approximately 248 individuals. As such, it appears that the population has moderate genetic diversity, but is also accumulating relatedness because of its intense selection. In contrast to the pedigree-based estimate of inbreeding, the genomic estimate using runs of homozygosity (ROH) indicated much higher inbreeding (mean $F_{ROH} = 0.0905$). Therefore, it appears that pedigree-based estimates of inbreeding are lower than true amounts of autozygosity. Further investigation into the distribution of ROH segments illustrated that nearly all genomic inbreeding occurred from short and intermediate ROHs. Since these ROH segments reflect largely old genetic relationship, relatively little evidence exists to suggest new or current inbreeding. Overall, it is evident that pedigree alone does not accurately depict the amount of inbreeding in modern dairy populations. By combining pedigree with genomic information, a more complete picture of both the structural makeup and history of a dairy population can be gained. Furthermore, utilizing combined pedigree and genomic information will provide dairy producers with a valuable resource when attempting to manage the long-term sustainability of genetic diversity within Holstein cattle.

Keywords: Holstein cattle, pedigree analysis, inbreeding, runs of homozygosity (ROH, genomic inbreeding)

Cage shellfish aquaculture: the future of sustainable mussel farming?

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ABSTRACT

Mussel *Mytilus galloprovincialis* aquaculture along the eastern Adriatic coast has experienced a severe decline, with total production dropping from approximately 3,000 t in 2012 to around 938 t in 2025. This collapse is mainly due to increasing predation by gilthead seabream *Sparus aurata*, a species whose population has grown as a result of climate-driven sea surface warming, escapes from mariculture facilities, and abundant food at shellfish farms, with predation losses reaching up to 90% of total yield. The severity of these losses has prompted the Croatian Ministry of Agriculture, Forestry and Fisheries to compensate farmers for predator-induced damages. Although the net system provides mechanical protection and maintains yield, it fails to preserve product quality: dense biofouling on the netting and the close proximity of the mesh to the longline impair water circulation, restrict mussel feeding and ultimately compromise growth rates and final product quality.

The project “Sustainable Mussel Farming under Predator Pressure” aims to evaluate a novel cage-based protection system as a more efficient, economically viable and quality-preserving alternative. Three cultivation approaches will be compared at concession sites in Split-Dalmatia County: traditional rope cultivation, the net system, and the proposed cage concept. Abiotic and biotic parameters will be continuously monitored, along with assessments of yield, growth, nutritional quality, and cost-benefit performance.

Cage aquaculture is expected to significantly reduce predation losses while maintaining both the quantity and quality of mussels, providing a scalable solution for Croatian shellfish farmers in accordance with the National Strategic Plan for Aquaculture Development 2022–2027. This project is funded and supported by HAMAG-BICRO under the PoC programme, with support from the Ministry of Science, Education and Youth.

Keywords: mussel aquaculture, predator protection, *Sparus aurata*, cage farming, Adriatic Sea

Assessment of milk composition and selected blood biochemical parameters in imported Lacaune sheep under production conditions in North Macedonia

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ABSTRACT

The aim of this study was to analyze the milk composition and selected serum biochemical parameters in Lacaune ewes in the final stage of lactation, in order to evaluate their productive and metabolic response to the diet in the conditions of North Macedonia. Milk samples were collected from ewes in their third lactation during three consecutive months, namely July (n = 29), August (n = 26) and September (n = 22), while blood samples were collected and analyzed only once, in September (n = 39). The diet consisted of concentrate, alfalfa hay and clover. Milk was analyzed for total bacterial count (TBC), somatic cell count (SCC), fat, protein, lactose, solids-not-fat (SNF) and total dry matter, while serum biochemical analysis included indicators of liver function, energy metabolism, protein metabolism and mineral status. The results showed that during late lactation, fat, protein, SNF and total dry matter increased, reaching 9.57%, 9.41%, 13.47% and 24.09%, respectively, whereas lactose decreased to 3.36%. Mean TBC increased from 225,241.38 CFU/mL in July to 794,454.55 CFU/mL in September, while mean SCC increased from 161,793.10 to 649,772.73 cells/mL. Differences between July and September, as well as between August and September, were statistically significant for all examined milk parameters ($p < 0.001$), whereas no significant differences between July and August were found for TBC, SCC and fat content ($p > 0.05$). Monthly comparisons were performed using the independent samples t-test with Welch correction. The serum biochemical profile indicated a relatively stable metabolic status in the examined animals, with no clearly expressed statistically significant relationships between the analyzed serum parameters and the main chemical components of milk. In conclusion, the results suggest that Lacaune ewes showed satisfactory productive and metabolic response under the given conditions, and that the observed changes in milk composition were consistent with the physiological pattern of late lactation.

Keywords: Lacaune ewes, milk composition, late lactation, blood biochemical parameters.

Polymer composition and size distribution of microplastics in commercial animal feed samples

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ABSTRACT

Microplastics in animal feed are considered a potential pathway for the transfer of plastic particles into the food chain. This study aimed to characterize the polymer composition and particle size distribution of microplastics in commercial pig and poultry feed available on the Croatian market. A total of 16 feed samples from five producers were analyzed, including pig ($n = 10$) and poultry ($n = 6$) feed. Samples underwent oxidative digestion to remove organic matter, followed by filtration and polymer identification using FTIR microscopy (Lumos II equipped with an FPA detector). Various polymer types were identified, including polyethylene (PE), polypropylene (PP), polyester (PET), polyamide (PA), polyvinyl chloride (PVC), chlorinated polyethylene, rubber-based materials, and acrylates/polyurethanes. Particle size was expressed as projected area (μm^2). Differences between pig and poultry feed were assessed using the Mann–Whitney U test for each polymer type. A statistically significant larger particle size in pig feed compared to poultry feed was observed for chlorinated polyethylene ($P = 0.0197$), while polypropylene exhibited a near-significant trend ($P = 0.062$), larger in pig feed, too. No significant differences were found for the remaining polymers ($P > 0.05$). Variability in particle size and limited sample numbers for certain polymers may have affected the statistical outcomes. The presence of PVC, chlorinated polyethylene, and acrylate/polyurethane-based materials points toward additional contamination from industrial infrastructure, such as pipes, coatings, and equipment surfaces. The predominance of PP, which are widely used in packaging materials and agricultural applications, indicates that contamination likely occurs during feed processing, storage, and packaging, as well as from environmental exposure of raw materials. Overall, the results indicate that microplastics are consistently present in commercial pig and poultry feed, with similar polymer profiles and particle size distributions.

Keywords: microplastic, animal feed, FTIR microscopy, polymer composition, particle size

Acknowledgment: The research is funded by the European Union - NextGenerationEU (NRRP project „Nutritional modelling and metabolomics in the function of improving livestock production “ HRAMSTOC, no. 581-UNIOS 26”).

Physicochemical, spectral and melissopalynological characterisation of Croatian buckwheat (*Fagopyrum esculentum*) honey

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ABSTRACT

Buckwheat honey (BWH) stands out among European unifloral honeys due to its dark colour, unique organoleptic profile, high antioxidant activity, as well as high phenolic and mineral content. In Croatia, buckwheat (*Fagopyrum esculentum*) is cultivated on only ~500 ha, mainly in the Western Pannonian region, resulting in limited production and scarce research of BWH. Therefore, the aim of this study was to determine physicochemical, spectral and melissopalynological properties of rare Croatian BWH. A total of 10 BWH samples were collected directly from beekeepers across the Western Pannonian region of Croatia between 2018 and 2025. Samples were analysed by physicochemical analyses (determination of water content-WC, electrical conductivity-EC, and pH value), Fourier transform infrared spectroscopy coupled with Attenuated Total Reflectance (FTIR-ATR), and qualitative melissopalynological analysis. The results of physicochemical analyses of BWH samples showed mean (min–max) WC, EC, and pH values of 16.7% (14.6–19.2%), 0.37 mS/cm (0.22–0.42 mS/cm), and 3.82 (3.71–3.92), respectively. FTIR-ATR profiling revealed similar spectral patterns of analysed BWH samples indicating similar overall chemical composition, while melissopalynological analysis confirmed dominance of *F. esculentum* pollen grains in all samples, with percentages ranging from 20% to 56%. The pollen grains of other frequently represented accompanying nectariferous plant species, *Hedera helix*, Brassicaceae, *Solidago canadensis*, and *Tilia* spp. were also determined thus reflecting botanical environment of buckwheat cultivation areas in Western Pannonian region. Chestnut (*Castanea sativa*) pollen was also determined, but as a result of secondary and/or tertiary enrichment. A requirement for uniflorality on the national level (Ordinance on the quality of unifloral honey, Official Gazette NN 122/09) defined as at least 45% of pollen grains of decisive plant species, was not met for most of analysed samples. Moreover, 7 out of 10 samples contained <45% of *F. esculentum* pollen but still showed typical BWH profiles. These findings, together with available literature data, question the appropriateness of the current pollen percentage thresholds for BWH and suggest that national legislation should be revised, including the consideration of lowering the required minimal pollen grain threshold to 20%.

Keywords: buckwheat honey, physicochemical characterisation, FTIR-ATR spectral profiles, melissopalynological properties

Acknowledgement: This research was funded by Research and Development of Plant Genetic Resources for Sustainable Agriculture, Center of Excellence for Biodiversity and Molecular Plant Breeding (CoE CroP-Bio-Div), Zagreb, Croatia (PK.1.1.10.0008).

Session 3

Agroecology and Regenerative Agriculture

Investigation of the effects of regenerative farming systems on the intensity of soil respiration

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ABSTRACT

Soil organic matter is the foundation of soil health and fertility, promoting carbon sequestration, nutrient cycling, and structural stability. However, as a result of intensive cropping and climate change, a significant decrease in the organic matter content of Europe's soils has been observed in recent decades. Therefore, sustainable soil management practices should be applied to accumulate soil organic matter, maintain soil fertility and mitigate the adverse effects of climate change. To better understand the effect of soil processes on soil health, a tillage crop rotation experiment was established with a variety of green manure species and their mixtures in 2024 at the Research Institute of Nyíregyháza IAREF University of Debrecen. The aim of the research was to assess the impact of conventional and regenerative farming practices on carbon emissions of sandy soil (Arenosol). Soil respiration measurements in the field were done twice during the growing season in 2025 by a portable device of BIOBASE SRM-3051T. Soil respiration showed significant variance across tillage treatments and crop types. In winter wheat and green peas, the intensity of soil respiration was generally higher in conventional plots compared to regenerative treatments. However, in the case of maize, the highest rates of soil CO₂ emissions were generally observed in regenerative plots. In conclusion, the significant differences in soil respiration intensity between treatments confirm the impact of different tillage practices on soil biological activity, however, long-term measurements are needed to establish definitive trends regarding carbon sequestration.

Keywords: regenerative farming; green manure; soil health; sandy soil; soil respiration

The Influence of Treatment Area Size on the Efficacy of Mating Disruption for the Management of *Cydalima perspectalis* (Lepidoptera: Crambidae)

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ABSTRACT

The aim of this study was to evaluate the applicability of mating disruption (MD) and to determine the influence of treatment area size on its performance in controlling the invasive box tree moth (*Cydalima perspectalis*) in urban landscapes. Field experiments were conducted in Split-Dalmatia County (Croatia) during 2023 in urban areas with infested boxwood plants. CIDETRAK[®] BTM dispensers were applied at a density of 50 dispensers ha⁻¹ across plots ranging from 0.02 to 4 ha in a randomized complete block design. The efficacy of MD was assessed by monitoring male moth captures using pheromone traps and by evaluating infestation levels and plant damage through sampling of existing and sentinel plants. Preliminary results indicate a reduction in male moth activity in treated plots and a decreasing trend in infestation levels compared to untreated areas. The results also suggest that treatment area size affects the performance of MD, with larger plots showing more consistent effects, while smaller plots exhibit greater variability. In conclusion, mating disruption shows potential as an environmentally acceptable method for managing *C. perspectalis* in urban environments.

Keywords: *Cydalima perspectalis*, mating disruption, urban pest management, pheromones, boxwood

Life Cycle Assessment of Innovative Biowaste-Based Hydroponic Substrates Using the Cut-off and APOS System Models

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ABSTRACT

Valorisation of agricultural waste materials for hydroponic substrate production represents a promising approach to reducing environmental burden within the circular economy framework. This study evaluates the environmental impacts of an innovative bio-hydroponic substrate formulated from washed sheep wool fibres, biochar from wood-chip pyrolysis, biomass fly ash, and walnut husk using the Life Cycle Assessment (LCA) methodology. Two substrate formulations with varying mass fractions were modelled in SimaPro 9.1.1.1 using the ReCiPe 2016 Midpoint (H) method with 1 kg of substrate as the functional unit within gate-to-gate system boundaries. The Ecoinvent 3.6 background database was regionally adjusted to represent Croatian extensive sheep husbandry practices, as global default assumptions on feed inputs, pasture management, and water use significantly overestimate environmental impacts under local conditions. Both the cut-off and Allocation at Point of Substitution (APOS) inventory methods were applied. Under the cut-off method, wool, fly ash, and walnut husk carry no upstream burden as waste-derived materials. Impacts identified under the APOS method are partly attributable to the allocation of sheep farming burdens to wool as a co-product, with copper emissions from upstream livestock housing construction as the dominant contributor to ecotoxic impacts on aquatic ecosystems and human health. The lower-wool formulation exhibited a lower ecotoxicity footprint compared to the higher-wool formulation, with wool washing and drying identified as the principal burden contributor under the cut-off method. Biochar application contributes approximately 2.7 kg CO₂ kg⁻¹ of net carbon sequestration, representing a significant climate co-benefit. These findings confirm that bio-hydroponic substrates based on the valorisation of domestic agricultural waste represent an environmentally justified circular economy approach, with further improvement potential through energy decarbonisation.

Keywords: life cycle assessment, bio-hydroponic substrate, sheep wool, biochar, fly ash, walnut husk

Physiology and biochemistry of different chickpea varieties as affected by late sowing under supplemental irrigation

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ABSTRACT

Chickpea is not cultivated in Hungary nowadays. However, the engagement of chickpea in a wheat-chickpea crop rotation to guarantee a sustainable, all-year utilization of arable lands seems feasible, but the physiological and biochemical responses of chickpea to late sowing, and the possible role of supplemental irrigation are not evaluated yet in Hungary. An experiment was conducted at the experimental farm of the Faculty of Agricultural and Food Sciences and Environmental Management of the University of Debrecen in 2024 and 2025. Three chickpea varieties (Amorgos, Elmo, and Orion) were sown in July (after wheat harvesting) and supplemental irrigation was applied and compared to non-irrigated scheme. The number of days to flowering (DTF) of the non-irrigated plants of all varieties was less compared to their irrigated counterparts. DTF of Orion was significantly higher in comparison with the other varieties under the non-irrigated scheme. Irrigation did not have any effect on the normalized difference vegetation index (NDVI) and Elmo had the highest NDVI among the tested varieties. The total carotenoid content of all varieties was higher under non-irrigated, compared to irrigated scheme. Under the non-irrigated scheme, the total carotenoid content increased significantly in Elmo but decreased significantly in Orion. Furthermore, there were no significant differences in the phenolic, tannin and flavonoid contents, and the antioxidant activity in the seeds harvested from the irrigated plots as compared to their non-irrigated counterparts. It could be concluded that irrigation does not seem to influence the physiology or the biochemistry of chickpea sown after wheat harvesting in Hungary. However, yield components and seed quality traits must be evaluated under such conditions. Moreover, selecting suitable varieties is of high importance.

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Keywords: flavonoids, chickpea, phenols, total carotenoids

Innovations for Blue–Green Bioeconomy Integration in Circular Agrifood Systems

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ABSTRACT

Global food systems are increasingly challenged by climate change, nutrient pollution, and inefficient resource use, while agriculture and aquaculture remain largely separated despite their potential for circular nutrient exchange. The objective of this study is to identify and analyse innovations that enable the integration of blue and green bioeconomy within circular agrifood systems and to examine their contribution to systemic transformation of food production. A systematic literature review was conducted using a structured search string in the Web of Science database, focusing on publications from 2015–2025. The search yielded 593 publications, of which 260 were screened in detail, resulting in 151 papers containing at least one identifiable innovation. Among these, 33 papers addressed innovations connecting both blue and green bioeconomy systems, while 63 focused on blue bioeconomy innovations and 55 on green bioeconomy innovations. Cross-sectoral innovations identified in the literature include integrated aquaculture–agriculture systems such as aquaponics and rice–fish production, circular nutrient management approaches including wastewater reuse and nutrient recovery, biomass valorisation and alternative feed resources, and systemic framework innovations such as digital decision-support systems and nutrient flow analysis. Despite the growing number of sectoral innovations, the results reveal a clear imbalance, with relatively few studies explicitly addressing integrated blue–green bioeconomy solutions. This highlights a critical research and policy gap and indicates that advancing circular agrifood systems requires stronger cross-sectoral innovation frameworks and policy alignment to connect aquatic and terrestrial production systems.

Keywords: blue bioeconomy, green bioeconomy, circular agrifood systems, systemic innovation, integrated aquaculture–agriculture systems

Integration of phytoremediation, carbon sequestration and energy production by energy crops cultivation

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ABSTRACT

Soil contaminated with heavy metals will be delivered to the University of Zagreb, Faculty of Agriculture, where two energy crops (miscanthus/giant reed) will be cultivated in ex-situ conditions for 3 years. The possibility of soil remediation by phytoremediation will be investigated, as well as climate change mitigation through the use of solid biofuels, increasing carbon sinks (sequestration in biomass and soil) and reducing carbon sources (soil and plant respiration). The specific objectives will be: (1) Identify potential types of soil pollution and phytoremediation possibilities with the investigated crops/soil improver; (2) Determine biomass growth and biological and economic yield; (3) Determine the energy properties of biomass and evaluate the impact of pollution on the use of biomass in the production of solid biofuels; (4) Determine the impact of various pedo-climatic factors, types of energy crops and the application of soil improvers on carbon cycling (sources and sinks) in the soil-plant-atmosphere system; (5) Develop models and calculate the biological, economic, phytoremediation, energy potential as well as biological carbon sequestration of the studied crops using machine learning approaches. To achieve the goals, the following will be carried out: (a) physical, chemical and elemental soil analyses and identification of the presence and type of pollutants; (b) elemental composition analyses of belowground biomass; (c) elemental and energy analyses of aboveground biomass; (d) monitoring of agroecological conditions and carbon cycling at 1 ex-situ and 2 in-situ locations. New findings will contribute to a better understanding of soil remediation by phytoremediation, bioenergy production, as well as the complexity of the interaction of agroecosystem factors on carbon flows.

Keywords: phytoremediation, energy crops, carbon sequestration, contaminated soil, solid biofuels

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Carbon sequestration options across Croatian agricultural land use

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ABSTRACT

Croatian agriculture is still dominated by cultivation practices that accelerate carbon losses, but the national pedosphere also offers considerable potential for carbon sequestration if management is adapted to land use and environmental conditions. The aim of this paper was to synthesize current management on annual croplands, perennial croplands and grasslands in Croatia and identify the most relevant measures for increasing soil carbon stocks. The analysis considered the three main agricultural regions of Croatia, the Pannonian, Mountainous and Adriatic. The review showed that annual croplands are mainly managed conventionally, with more than 90% of farmers using invertive tillage, while conservation systems occupy less than 10% of arable land. Extra constraints include simplified rotations, scarce cover crops, improper fertilization, stubble or pasture burning and poorly managed grazing, all of which favour carbon loss and soil degradation. Across land uses, the strongest opportunities for improvement were linked to conservation tillage, diversified crop rotations with legumes and perennial forages, cover crops, optimized organic and mineral fertilization, permanent grass cover in perennial crops, agroforestry and silvopastoral systems. The suitability of each measure depends on soil type, climate and regional conditions, so a uniform solution on national level cannot be applied. The synthesis indicates that Croatian agricultural soils still have considerable capacity to sequester carbon, but this potential will remain underused without stronger incentives, field testing and clearer implementation guidelines. Site-specific support measures and policy alignment are therefore needed to turn carbon farming from a strategic objective into routine practice.

Keywords: carbon sequestration, carbon farming, croplands, grasslands, Croatia

Acknowledgment: This work was supported by the Croatian Science Foundation through the “Forming climate-smart soils: Mitigation of soil erosion and degradation processes in Croatian agricultural systems” project (IP-2022-10-5692) (FORMclimaSOIL).

Conservation tillage reduces erosion in a sloping maize–wheat agroforestry system

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ABSTRACT

Soil erosion is a major threat in hilly croplands of Croatia, especially where conventional tillage is applied up and down the slope. This study evaluated whether conservation tillage can improve soil health and reduce overland flow in a young plum orchard with a maize–wheat rotation established in interrows on a sloping Stagnosol in northwestern Croatia. Paired plots (20 m², 3 replicates, 6 in total) were used to compare conventional tillage (ploughing and discing) and conservation tillage (loosening and harrowing) during two crop seasons under natural rainfall. Soil physical and chemical properties were monitored, along with runoff, sediment concentration, soil loss, and associated element export. Conservation tillage resulted in higher aggregate stability and higher soil carbon at the 0–10 cm depth, while no clear increase in compaction was recorded during the transition period. At the same time, hydrological and erosional responses were markedly reduced. Mean sediment concentration decreased from 8.95 to 3.11 g L⁻¹, runoff was 40.5% lower, and cumulative soil loss declined from 8.57 to 2.97 t ha⁻¹. Lower sediment export also reduced off-site losses of nutrients and copper; compared with conservation tillage, conventional tillage increased P losses about 3.4-fold, K losses 3.2-fold, Cu losses 2.6-fold and N losses 1.7-fold. The results indicate that non-inversion conservation tillage is a practical climate-smart option for sloping cereal production in orchard inter-rows because it simultaneously reduces erosion risk, limits pollutant transfer and supports soil functionality.

Keywords: conservation tillage, soil erosion, runoff, Stagnosol, maize–wheat rotation

Acknowledgment: This work was supported by the Croatian Science Foundation through the “Forming climate-smart soils: Mitigation of soil erosion and degradation processes in Croatian agricultural systems” project (IP-2022-10-5692) (FORMclimaSOIL).

Smarter Coatings, Stronger Strawberries

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ABSTRACT

This study investigated the application of edible coatings on strawberries formulated from carboxymethyl cellulose and polyvinyl alcohol, with the incorporation of grape pomace extract as a natural bioactive additive. The aim was to evaluate the effectiveness of these coatings in preserving the antioxidant properties of strawberries during 11 days of cold storage. Antioxidant potential was determined spectrophotometrically in strawberry juice using two assays, DPPH and ABTS, which measure radical scavenging activity. The results demonstrated that strawberries coated with edible films retained a higher level of antioxidant capacity compared to uncoated control samples, indicating a protective effect of coatings against oxidative degradation. Furthermore, the incorporation of grape pomace extract into the coating formulation resulted in a slight but consistent increase in antioxidant activity, likely due to the presence of phenolic compounds with known antioxidative properties. Overall, results of this experiment suggest that these biodegradable edible coatings not only act as a physical barrier but also contribute functionally to the preservation of bioactive compounds. Therefore, they show promising potential as an innovative approach for extending shelf life and maintaining the nutritional and functional quality of fresh strawberries.

Keywords: edible coatings, strawberry, antioxidant potential, postharvest

Heat sensitivity index in maize across different maturity groups

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ABSTRACT

Maize is one of the most important crops in the world and climate changes affect its production worldwide. Various scenarios of abiotic stresses have emerged – some of the harshest being heat and drought – increasing time and effort of plant breeders and farmers who must address new challenges in a limited space-time frame. Earlier maturity hybrids with shorter vegetation could have an advantage in maize production (coping with or avoiding heat stress in pollination and other vegetation stages). Heat sensitivity index (HSI) is a fluorescence parameter for evaluating the plant's response to high temperatures. It shows the relation between the logarithm of performance index ($\log PI_{ABS}$) and relative variable fluorescence at the K-step (V_K) of the fluorescence transient curve. Generally, lower HSI scores suggest higher tolerance to heat stress. The aim of this study was to assess the effect of high temperatures on maize hybrids from various maturity groups using HSI. The lowest HSI scores (-12.84 and -12.28) were recorded in grain filling stage of earlier maturity groups (FAO 200 and FAO 400) while late hybrids from FAO 700 maturity group showed highest HSI scores in both anthesis (-6.28) and grain filling (-3.96). Across maturity groups and vegetation stages, HSI scores were lowest in grain filling in all groups except FAO 700. The lowest average HSI values across all vegetative stages were recorded in hybrids from FAO 400 maturity groups, while the highest average HSI values were recorded in hybrids from FAO 700 maturity group. These results show that late materials can be very sensitive to heat stress, while selecting hybrids from FAO 400 could be a valuable strategy. More research with different parameters is required in order to get wider picture about the complex problem of heat stress.

Keywords: maize, maturity, abiotic stress, heat sensitivity index

Biodegradable microplastics and soil quality

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ABSTRACT

Biodegradable plastics are increasingly promoted as a sustainable alternative to conventional plastics, with the potential to reduce the accumulation of persistent microplastics in soils. However, their rapid degradation and interactions with soil processes raise important questions regarding their effects on soil quality and ecosystem functioning. In this context, we investigated the behavior of poly-3-hydroxybutyrate (P3HB), a biodegradable polymer of microbial origin, and its impact on soil properties and plant performance across a gradient of concentrations relevant to agricultural applications and plastic contamination scenarios.

Our results demonstrate that P3HB strongly influences soil quality by altering nutrient dynamics, microbial activity, and soil physical conditions. At higher concentrations, P3HB negatively affected plant growth and nutritional status, primarily due to imbalances in nutrient availability and shifts in the soil microbial community. Enhanced microbial activity during P3HB degradation led to increased enzyme production and stimulated soil organic matter turnover, indicating a priming-like response. This process was accompanied by high microbial carbon utilization and was further modulated by soil texture, with rapid biodegradation leading to oxygen depletion and a transition from aerobic to anaerobic conditions. Attempts to mitigate these effects using compost or plant growth-promoting bacteria were ineffective, while digestate provided only limited improvement in plant performance.

Despite these negative effects, P3HB also exhibited context-dependent benefits. As a readily degradable carbon source, it can temporarily enhance microbial activity and contribute to short-term increases in soil organic matter, particularly in degraded soils. Overall, our findings highlight that biodegradable microplastics represent a double-edged factor for soil quality, with outcomes depending on their concentration, environmental conditions, and soil properties. These results provide a basis for the responsible use of biodegradable plastics in agriculture and soil management, emphasizing the need to balance their potential benefits with risks to soil functioning.

Keywords: biodegradable plastics; microbial processes; nutrient imbalance; plant–soil interactions; ecosystem functioning

Stakeholder perspectives on water reuse in agri-food and livestock systems

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ABSTRACT

As global climate change intensifies water scarcity, the agricultural sector responsible for up to 90% of freshwater consumption urgently requires sustainable alternatives. Reclaimed wastewater is a viable solution. However, its implementation depends heavily on stakeholder acceptance. This study aims to analyze the diverse perspectives of farmers, consumers, policy actors, and health professionals regarding water reuse in agri-food and livestock systems.

Research results indicate that farmers approach water reuse with "cautious optimism," driven by water security needs but limited by concerns over soil fertility and market rejection. Economic incentives remain a primary facilitator for adoption. Conversely, consumer acceptance is frequently hindered by the "yuck factor," though trust in regulatory certification and indirect usage (e.g., non-food crops) mitigates this resistance. Public health and veterinary professionals maintain the most cautious stance, advocating for a "multi-barrier" approach to address risks from pathogens and emerging contaminants like microplastics. Comparison of international case studies reveals that transparency and early stakeholder involvement are critical for project success.

In conclusion, while technical solutions for water reclamation are advanced, social and institutional barriers remain significant. Successful integration of reclaimed water in agriculture requires a shift from top-down management to inclusive, trust-based frameworks that address the specific economic and safety concerns of all stakeholders.

Keywords: reclaimed water, stakeholder acceptance, water scarcity, sustainable agriculture, food safety

Can green roofs save the city of Zagreb from flooding?

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ABSTRACT

Climate change and rapid urbanization have led to increasingly frequent extreme rainfall events, significantly raising the risk of urban flooding and surface runoff accumulation. The objective of this study was to evaluate the hydrological efficiency of green roofs in mitigating urban runoff and peak flows within a densely built-up area of Zagreb, Croatia. Using the Storm Water Management Model (SWMM), three green roof coverage scenarios were analyzed: a reference scenario (S1, 0%), moderate implementation on residential buildings (S2, 45%), and maximum implementation on all suitable flat roofs (S3, 65%). The model was tested against three rainfall scenarios: a continuous five-year series (O1) for long-term efficiency, an extreme event (O2) for peak flow reduction, and multi-day episodes (O3) for retention capacity and vegetation dynamics under saturated conditions. Results from O2 showed that while S1 resulted in a runoff coefficient of 0.99, S3 proved most effective, reducing total runoff by 45% and halving the peak flow (50% reduction). Long-term simulations (O1) confirmed consistent volume reduction across different years, while O3 highlighted the importance of seasonal vegetation processes in restoring retention capacity between successive events. Even under repeated rainfall, green roofs demonstrated substantial cumulative retention, particularly during the growing season. It is concluded that while S2 provides initial benefits, S3 offers the highest potential for enhancing urban flood resilience. These findings emphasize the importance of integrating green infrastructure into urban planning to mitigate climate change risks and support the long-term sustainability of cities by transforming rooftops into functional ecological assets.

Keywords: green roofs, urban runoff, SWMM modeling, peak flow reduction, sustainability

Chemical Characterization of Sheep Wool from Croatian Breeds by FTIR-ATR Spectroscopy

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ABSTRACT

Sheep wool represents a significant agricultural by-product in Croatia, generated through regular shearing practices. Although sheep wool is a renewable and biodegradable material rich in organic nitrogen and sulphur, substantial quantities remain underutilized and are often disposed as waste. Its potential application within agroecosystems largely depends on its physicochemical properties, which may vary depending on the sheep breed and breeding location. The aim of this study was to determine the biochemical composition of sheep wool from two Croatian breed groups using Fourier transform infrared spectroscopy coupled with attenuated total reflectance (FTIR-ATR).

Two types of wool originating from Dalmatian pramenka and Pramenka type of sheep, representing different Croatian regions, were analysed in the mid-infrared region (4000–600 cm^{-1}). The results of spectral analysis revealed that IR spectra of analysed wool samples were dominated by keratin structure, fatty acids, esters, and lanolin content. The results exhibited notable spectral variations between analysed sheep wool breeds, in terms of both the position and intensity of absorption bands thus reflecting compositional variations in investigated wool samples. Dalmatian pramenka wool exhibited consistently stronger absorption bands. A characteristic band at 1737 cm^{-1} , attributed to C=O stretching vibrations of monoesters, including saturated aliphatic esters and free fatty acids, further confirms the presence of lanolin and was observed exclusively in the Dalmatian pramenka wool. Additionally, a weak band at 874 cm^{-1} was detected only in Dalmatian pramenka wool and may be assigned to C–C skeletal vibrations associated with the keratin backbone.

The obtained results demonstrate that FTIR-ATR spectroscopy is a reliable tool for rapid chemical characterization of sheep wool at the molecular level. Further research should focus on correlating the spectral features of sheep wool with nutrient release dynamics and soil improvement potential.

Keywords: sheep wool, FTIR-ATR spectroscopy, chemical composition, sustainable agriculture by-product

The possibilities of application sheep wool in bell pepper production

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ABSTRACT

The aim of this study was: i) to determine the effect of applying different dead organic (wool) mulches (raw wool – SW, "felted" wool – FW), polyethylene film (PE), and unmulched soil (WM) on soil moisture content (SMC) and the yield of bell pepper (*Capsicum annuum L., ssp. Macrocarpum, var. Blanchina FI*) during the growing season (May–September, 2017); ii) to analyse the possibility of using wool materials as an ecologically and economically acceptable substitute for PE materials in pepper production. The research was conducted at the Faculty of Agronomy in Zagreb, and the experiment was set up according to a randomised block design with four replications. Soil samples for gravimetric determination of SMC were taken every ten days from the surface layer (0–10 cm) and the subsurface layer (10–20 cm), in three replications from each mulch treatment. The days after transplanting the pepper outdoors (DAT) and the type of mulch (M) had a statistically significant effect on SMC in both soil layers. In the 0–10 cm soil layer, SMC varied between 29.61% vol. (95 DAT) and 38.14% vol. (15 DAT), and in the 10–20 cm layer between 28.63% vol. (95 DAT) and 38.34% vol. (15 DAT). Regarding the type of mulch in both soil layers, a significant difference in soil moisture was found between the wool treatments (SW and FW) compared to WM and PE. The lowest SMC was recorded under PE (31.23% vol. in the surface and 31.01% vol. in the subsurface layer), and the highest SMC under SW (35.04% vol.) in the surface, and under FW (35.41% vol.) in the subsurface layer. The highest total pepper yield (0.86 kg/m²) was recorded on August 3, and the lowest (0.24 kg/m²) was recorded on September 4. Regarding M, the highest total pepper yield was achieved on SW (0.69 kg/m²), and the lowest on PE (0.43 kg/m²), with no statistically significant differences in yield between WM, FW, and PE. The application of raw wool in this experiment ensured optimal microclimatic conditions in the soil necessary for pepper cultivation. Although sheep wool has been considered waste, thanks to its thermoregulatory properties and high biodegradability, sheep wool has great ecological and economic potential for use not only in vegetable growing, but also in other branches of agriculture.

Key words: bell pepper, soil moisture content, wool mulch, yield.

Novel encapsulation of endophytic bacteria and *Trichoderma* against *Macrophomina* in beans

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ABSTRACT

Dry root rot caused by *Macrophomina phaseolina* threatens bush bean production in India and Southeast Asia, with disease incidence exceeding 50% under heat and moisture stress. This study aimed to develop a novel biological control agent (BCA) using endophytic microorganisms as a sustainable alternative to chemical fungicides. From healthy host plants (pineapple, palm, mango, citrus grass, neem seed), 221 endophytic isolates were obtained. Initial in vitro dual culture screening against *Macrophomina* identified 3-4 bacterial isolates showing 60-75% mycelial growth inhibition. Planned methodology includes molecular identification, GC-MS analysis of volatile organic compounds, whole-genome sequencing to identify biosynthetic gene clusters, synergy testing with *Trichoderma*, and formulation into a stable capsule. Results confirmed strong antagonistic potential (inhibition up to 75%). Genomic and metabolomic analyses will reveal genetic determinants of biocontrol activity. The capsule formulation is designed to improve viability and shelf-life for practical farm use. We conclude that this integrated pipeline from endophyte isolation to product development offers a resilient, smart-agriculture solution for managing *Macrophomina* root rot, translating research into an accessible grower tool.

Keywords: Biological control, *Macrophomina phaseolina*, endophytes, *Trichoderma*, capsule formulation

Microplastics in the watercourse and coastal mud of the river Miljacka

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ABSTRACT

This study aimed to assess the degree of pollution by micro, meso and macro plastic waste and its sustainable solution and removal. Also, to implement a strategic plan to discover, collect, determined by size and type of collected plastic and remove as much plastic as it possible from the Miljacka river from its spring to the estuary with the Bosna River.

The activities in this experiment have been guided primarily by the idea of detecting macroplastics and microplastics using various methods, physically collecting, recording the river flow and its surroundings with a drone, sampling (different kind of nets) and differentiation of collected microplastics.

The taken water and mud samples analysis by Furier Infrared Spectroscopy (FTIR) method included particles with dimensions smaller than 500 µm and larger than 20 µm, due to the limitations of the pore size of the filters used. The result is presented as the number of particles per liter of sample. The collected data showed that four types of microplastics prevailed in the water samples. These are: polyethylene (PE), polypropylene (PP), polyamide (PA) and ethylene-vinyl-acetate (EVA). In the first group of water samples, the percentage of each particle was: PE 68.7%, PP 14.5%, PA 13.7% and EVA 3.1%. In the second group, the ratio was: PE 8.3%, PP 20%, PA 52.8% and EVA 18.9%. In the third group, the ratio was: PE 49.2%, PP 26.3%, PA 22.8% and EVA 1.7%.

The presence of 8 types of polymers was detected in the coastal mud samples. Of the detected particles, 43.6% belong to the smallest category of dimensions up to 25 µm, while 94.2% of the particles have dimensions less than 100 µm. The presence of larger particles was also detected, which make up the remaining 5.8% of the sample, with a maximum observed size of 350 µm.

Keywords: microplastic, water samples, coastal mud, determination, FTIR

Role of Earthworms in Enhancing Maize Productivity and Climate Resilience in Smallholder Farming Systems: An Agroecological Review

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ABSTRACT

Maize (*Zea mays L.*) is a globally important crop cultivated on more than 200 million hectares and represents a major source of food, feed, and income, particularly within smallholder farming systems in sub-Saharan Africa, South Asia, and Latin America. Sustaining maize productivity under increasing climatic variability requires management strategies that strengthen soil health and ecological resilience. Earthworms, recognized as ecosystem engineers, influence soil structure, organic matter dynamics, nutrient cycling, and water regulation through burrowing, casting, and residue processing activities. This review aims to synthesize evidence on the functional roles of earthworms in maize-based agroecosystems across diverse climatic regions, with emphasis on soil processes, agroecological management interactions, and implications for climate resilience. A narrative synthesis of the literature indicates that earthworm activity is frequently associated with improvements in soil aggregation, porosity, moisture dynamics, and nutrient availability, contributing to enhanced crop performance and yield stability under environmental stress. Practices such as reduced tillage, organic residue retention, and cover cropping consistently promote earthworm abundance and activity. Despite increasing research attention, long-term and region-specific evidence remains limited, particularly in tropical smallholder systems. The findings highlight the importance of integrating soil biological processes into maize management and underscore the need for further standardized, long-term, and context-specific investigations.

Keywords: earthworms, soil health, agroecology, smallholder systems, climate resilience

Bioash Additions Reshape the Rhizobiome in Metal-Contaminated Soil

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ABSTRACT

Bioash is a solid byproduct generated in large quantities during the combustion of wood biomass in bioenergy systems. It is increasingly considered a promising amendment for contaminated soils, yet its impact on rhizosphere microbial communities remains poorly resolved. The aim of this study was to evaluate how bioashes differing in origin and acid-neutralizing capacity, applied at different rates, influence the rhizosphere microbiome of *Lactuca sativa* L. grown in metal-contaminated soil. Rhizosphere bacterial communities were characterized by 16S rRNA gene amplicon sequencing and assessed through diversity metrics and taxonomic profiling.

Bioash addition consistently increased rhizosphere pH and electrical conductivity and reduced the bioavailability of Cd and Zn, confirming its liming and metal-immobilizing capacity. Alpha-diversity responses were limited, whereas bacterial community composition shifted significantly among treatments ($R^2 = 0.46$, $p = 0.009$). Multivariate analyses identified rhizosphere pH, salinity, exchangeable K, exchangeable Cd and mineral N as the main factors associated with microbial community restructuring. At the phylum level, bioash increased the relative abundance of Pseudomonadota, whereas Bacillota and Bacteroidota declined, particularly at the highest application rates. Differential abundance analysis further indicated that only a limited subset of taxa responded consistently to bioash addition.

These results demonstrate that bioash can function as a dual-purpose amendment in metal-contaminated soil by simultaneously reducing trace metal availability and reshaping rhizosphere bacterial communities. Its remediation potential is therefore considerable, but the pronounced dose-dependent microbial responses indicate that bioash type and application rate must be carefully optimized to balance soil improvement with microbial ecological stability.

Keywords: bioash, rhizosphere microbiome, metal-contaminated soil, 16S rRNA, soil remediation

Utilization of carbon-based amendments during manure maturation and their subsequent application in crop production

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ABSTRACT

Carbon-based amendments are increasingly used to improve manure stabilization and nutrient management prior to soil application; however, their combined effects remain insufficiently resolved. This study evaluated how amendment type (wood chips vs. biochar), biochar particle size (>2, 1–2, 0.25–1, 0.125, and <0.125 mm), and application pathway (co-maturation vs. post-maturation co-application) regulate manure transformation and subsequent soil functioning and maize growth. A factorial pot experiment was conducted using a Haplic Luvisol amended with cattle manure derived from deep-bedding systems (control vs. amended variants). Manure was either co-matured with amendments for 8 weeks under suboptimal conditions or amended immediately prior to soil application. Wood-chip reduced manure pH and altered C composition, while biochar effects were dependent on particle size and timing. The finest biochar fraction (<0.125 mm) induced the strongest changes during co-maturation, decreased pH and reduced mineral N and K availability relative to other fractions. Indeed, pre-fermentation amendment increased mineral nitrogen by up to 66% and NH₄⁺ by up to 76% relative to the control, whereas combined chip–biochar co-maturation reduced N_{min} by ~34%; in contrast, post-maturation biochar addition resulted in moderate increases in mineral N (+17–36%) and NH₄⁺ (+22–43%), indicating a shift in N transformation pathways depending on amendment timing. Following soil application, treatment responses diverged according to particle size and interaction pathway. Co-matured biochar fractions >2 and 1–2 mm and co-applied fractions 0.25–1 to <0.125 mm increased soil pH, total C and N, and microbial nutrient acquisition indices relative to the manure-only control. Enhanced enzyme activities (NAG, urease, phosphatase, ARS) indicated enhanced microbial processing. Despite these changes, maize aboveground biomass did not increase and was reduced in several treatments. Therefore, the results showed that manure agronomic value is governed primarily by stabilization pathway and amendment interactions rather than total nutrient content. Co-maturation enhanced microbial transformation, while co-application modified soil physicochemical conditions. Nevertheless, both pathways may induce microbial stoichiometric constraints that limit short-term nutrient transfer to plants.

Keywords: manure maturation, biochar, amendment, woodchip, stoichiometry

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Liming is among the most important steps in preserving soil fertility in Croatia

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ABSTRACT

The aim of this paper is to present the importance and urgency of liming considering chemical properties of soils in the Republic of Croatia, primarily with regard to soil acidity, soil organic matter (SOM) content and the level of available phosphorus in the soil. For this purpose, the results of the analysis of a total of 128,170 samples (total of 511,270 ha) of the arable soil layer were collected during soil fertility control in the period 2019-2024 and used to model the soil cation exchange capacity (CEC) and the degree of soil base saturation (BS). Based on pH value, SOM content, hydrolytic acidity, CEC and BS, the urgency of liming was evaluated in 6 levels: 1. very urgent, 2. urgent, 3. moderate urgency, 4. low need, 5. very low need for liming and 6. no liming required. A total of 49.51% of samples were acidic or very acidic ($\text{pH}_{\text{KCl}} < 5.5$), while $< 2\%$ SOM was in 39.02% of samples. Very low or low P availability was determined in 47.85% of samples, and K in 19.54% of samples. The model evaluated that liming was necessary or useful on 42.65% of the analyzed areas (218,039 ha) out of a total of 511,270 ha and not needed on 57.35% (293,231 ha). Liming was very urgently needed on 21,045 ha (4.12% of the area), urgently needed on another 66,933 ha (13.09%) and moderately urgent on 32,741 ha (6.40% of the area). In total, liming is urgently needed on 23.61% of the area (120,718 ha out of 511,270 ha). However, liming is also needed on an additional 31,655 ha (6.19% of the area), which makes up a total of 29.8% of the area (152,373 ha). The total required liming on the analyzed areas requires 1,762,960 t (8.1 t ha^{-1} in average) of pure limestone (CCE=100) or 2,518,514 t of carbocalc (CCE=70), which means 352,592 t of limestone per year over a five-year period. According to the urgency of liming, 237,775 t of limestone or 339,678 t of carbocalc are required annually over 5 years for the liming three the most degraded soil classes. Therefore, it's high time to launch a program of systematic soil liming, especially considering the degradation of SOM content and low P availability.

Keywords: soil degradation, SOM content, soil acidity, CEC, BS, urgency of liming

Climate change in Istria: analysis of elements and indexes

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ABSTRACT

Climate is a complex ecological factor which is described with different climatic elements, phenomena and indexes, which altogether have impact on development and natural distribution of vegetation. In terms of climate and vegetation, there are two vegetation zones in the Mediterranean-littoral vegetation belt of Istrian County. Those are: the sub-Mediterranean vegetation zone where the main forest tree species is the pubescent oak (*Quercus pubescens*), and the eu-Mediterranean vegetation zone, where the main forest tree species is the holm oak (*Quercus ilex*). The aim of the research was to analyze climate change in the Istrian region. For the analysis of the climate in the Istrian region, data from the Botonega and Pazin meteorological stations for the sub-Mediterranean vegetation zone and Poreč, Rovinj, and Pula meteorological stations for the eu-Mediterranean vegetation zone were used. The data were collected from the Croatian Meteorological and Hydrological Service for the period 1980–2022. Wind data were collected from the web platform KNMI Climate Explorer for the period 1980-2020 for the Pula area. Meteorological drought was analyzed using the rain anomaly index (RAI). Trends in mean annual air temperature show a significant increase, except for the Botonega meteorological station, while the trend in potential evapotranspiration had a significant increase for the entire Istrian region. Annual precipitation, length of the dry season (LDS), and the RAI index had an upward, but statistically insignificant trend. Average annual air temperatures increased significantly compared to the reference series, ranging from 0.8°C in Rovinj to 1.2°C in the Pula area. The southern Istria area had 3 dry months, which is characteristic of the steno-Mediterranean vegetation zone. Average wind speed and number of days with wind speed bigger than 10.8 m s⁻¹(strong wind which breaks tree branches) have statistically significant increasing trend. Data shows there are climate changes ongoing in Istria, which coincide with global climate changes trends.

Keywords: Istria, climate, vegetation zones

The impact of phenolic acids used as selective bioherbicides on beneficial soil microbiota

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ABSTRACT

Due to the harmful effects of synthetic herbicides on the environment and human health, as well as the rise of herbicide-resistant weeds, alternative weed control strategies are needed. Following the European Food Safety Authority's scientific opinion on the risk assessment of plant protection products for soil organisms, this study evaluated the minimum inhibitory (MIC) and minimum bactericidal concentrations (MBC) of selected phenolic acids (*p*-hydroxybenzoic (PHBA), protocatechuic (PKA), chlorogenic (CGA), ferulic (FA), vanillic (VA), and *p*-coumaric (PCA) acids) and their mixtures against different strains of beneficial soil bacteria from the genera *Pseudomonas*, *Bacillus*, *Peribacillus*, *Stenotrophomonas*, *Enterobacter*, *Lelliottia*, and *Bradyrhizobium*. The effect of subinhibitory concentrations on bacterial motility was also assessed to identify potential morphological damage. Results showed that bacterial susceptibility varied among strains; however, a consistent pattern emerged in which phenolic acid mixtures exhibited greater antimicrobial activity than individual compounds. PKA and CGA demonstrated the strongest activity (MIC 2.5–40 mM), PKA inhibited 78% and killed 74% of strains at ≤ 10 mM, while CGA inhibited 61% and killed 57%. PCA and VA were least effective, requiring ≥ 20 mM for complete inhibition. The mixture of all six phenolic acids (MIX6) was the most potent, showing inhibitory and bactericidal effects at ≤ 5 mM and affecting 74% of strains at 2.5 mM. The PPK mixture (PHBA+CGA+PKA) was also very effective (2.5–10 mM), while FVP (FA+VA+PCA) displayed the weakest activity, primarily at higher concentrations. At the genus level, susceptibility patterns were consistent with strain-level results, with PKA, CGA, PPK and MIX6 demonstrating the greatest efficacy. Subinhibitory concentrations did not affect bacterial motility, indicating no morphological damage. Given their low antibacterial activity at lower concentrations, vanillic, *p*-coumaric, and ferulic acids appear to be promising candidates for alternative weed control.

Keywords: weed control, minimum inhibitory (MIC) and minimum bactericidal concentrations (MBC), motility

Environmental suitability of using treated wastewater fertigation in loam soils of the Campania region (Southern Italy)

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ABSTRACT

The reuse of treated wastewater in agriculture through fertigation represents a viable strategy to reduce freshwater demand and improve nutrient circularity. Despite this, plant responses to irrigation with chemically distinct water types under specific soil conditions remain poorly characterized. This study assessed the effects of four synthetic irrigation waters (leachates, domestic wastewater, industrial effluent, and synthetic rainwater as control) on radish (*Raphanus sativus* L.) cultivated in loam soil from the Campania region under controlled pot conditions. Irrigation was conducted over approximately 40 days following CROPWAT scheduling (~10 L per treatment). Drainage water was analyzed for pH, electrical conductivity (EC), and ionic composition, while soil EC and moisture were continuously monitored. Plant performance was quantified through image-based measurements of leaf area and plant height. Results indicated that domestic wastewater and industrial effluent significantly enhanced plant growth compared with the control, whereas leachate treatments led to reduced performance. Elevated EC values in drainage were initially observed under nutrient-enriched treatments, reflecting increased ionic inputs and solute mobility, but reduced over time, suggesting an increase of nutrient uptake by plants. Plant yield and performance were strongly governed by water chemistry. These results show the importance of reused water composition in controlling nutrient availability, leaching dynamics, and crop response. This information is pivotal for further studies aimed at improving fertigation strategies.

Keywords: wastewater reuse, wastewater Fertigation feasibility, soil reaction, plant nutrient uptake, experimental scaling

Linking NDVI dynamics to crop yield under different cover crop and fertilization strategies in crop rotation systems

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ABSTRACT

Linking vegetation indices to crop performance provides a promising approach for evaluating the agronomic and ecological benefits of cover cropping systems in regenerative agriculture. This study investigated the effects of different cover crop species and mixtures, with or without supplementary mineral nitrogen, on NDVI dynamics and yield formation in maize and winter wheat.

The experiment was conducted on humic sandy soil in Hungary in a randomized block design with four replicates within a crop rotation system. Treatments included legume-based, brassica-based and mixed cover crops with or without supplementary mineral nitrogen, a mineral fertilization-only treatment (80 kg ha⁻¹ N), and an unfertilized control. NDVI was monitored throughout the growing season, and both peak NDVI and the area under the NDVI curve (AUC) were used to characterize cash crop development.

Cover crop effects were strongly crop-dependent. In maize, vetch-based treatments achieved yields comparable to mineral N fertilization, indicating their potential to partially substitute synthetic inputs. In contrast, in winter wheat, high yields were only obtained with additional mineral N application, suggesting that cover crops alone could not meet crop nutrient demand. The relationship between NDVI and yield also differed between crops. In maize, yield was more closely related to AUC than to peak NDVI, while in wheat both NDVI-derived metrics showed a strong association with yield. These results indicate that yield prediction based on a single NDVI measurement may be unreliable, especially in maize, whereas integrative indicators such as AUC provide a more robust representation of crop performance.

Overall, the findings highlight crop-specific responses to cover cropping and fertilization, and emphasize the importance of continuous NDVI monitoring for improving yield prediction and optimizing nutrient management in sustainable crop rotation systems.

Keywords: cover crops, NDVI dynamics, yield prediction, nitrogen management, crop rotation

Rare-Elements Dynamics (Nb, Zr, Y) in Zagreb's Peri-Urban Soils

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ABSTRACT

Peri-urban areas are dynamic environments where natural geochemical processes interact with anthropogenic influences. This study examines the seasonal and spatial variability of rare elements, niobium (Nb), zirconium (Zr), and yttrium (Y), in soils of Zagreb's peri-urban zone across five land-use types. Soil samples were collected in spring, summer, autumn, and winter of 2021 in Šašincev. Eight composite samples (0–30 cm) were taken from five distinct land-use: apple orchard, grassland, cropland, forest, and abandoned agricultural land. In total, 160 soil samples (8 per land use per season; 40 per sampling) were analyzed for elemental concentrations using a portable X-ray fluorescence analyzer. Ratios Nb/Zr and Y/Zr were calculated as indicators of geochemical processes and potential anthropogenic inputs. The two-way ANOVA and Tukey post hoc test results indicated that the accumulation of Nb, Zr and Y was affected by season and land uses. Regardless of land use or season, Y content ranged from 27.9 to 36.8 mg kg⁻¹, Zr from 220.4 to 415.4 mg kg⁻¹, and Nb from 17.5 to 27.4 mg/kg. Throughout the entire year, the Y content followed the sequence: cropland > apple orchard > grassland > abandoned agricultural land > forest, while Nb and Zr showed the opposite pattern: forest > grassland > cropland > apple orchard > abandoned agricultural land. Significantly the lowest Nb/Zr and Y/Zr ratios across all four seasons were recorded in the forest soil, ranging from 0.059 in autumn to 0.070 in winter for Nb/Zr, and from 0.080 in autumn to 0.085 in winter for Y/Zr, indicating the intense weathering and specific mineralogical composition in forest soils. In contrast, significantly the highest Nb/Zr and Y/Zr ratios were recorded in the apple orchard and the abandoned agricultural land. In autumn, summer, and spring, the highest Nb/Zr ratios (0.081–0.086) were detected in the orchard, while the highest Y/Zr ratios (0.144–0.148) were observed in autumn, spring, and winter on the abandoned agricultural land, reflecting a relative enrichment of Nb and Y compared to Zr. These elevated ratios likely result from differences in soil management and organic matter accumulation, which enhance the mobility and retention of Nb and Y in these land-use types. Over all, the findings reveal characteristic patterns in elemental distribution and contribute to a better understanding of rare-element dynamics in peri-urban soils, providing a basis for future monitoring and research.

Key words: soil geochemistry, Nb/Zr, Y/Zr, seasonal variability, peri-urban environment

Acknowledgments: This study has been funded by the Horizon Europe Project InBestSoil (GA 101091099) “Monetary valuation of soil ecosystem services and creation of initiatives to invest in soil health: setting a framework for the inclusion of soil health in business and in the policy making process”.

Allergenic plant species in Čapljina: Analysis of the pollen spectrum and impact on population health

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ABSTRACT

Allergenic plant species represent a significant source of aeroallergens that can cause various allergic reactions in susceptible individuals. Despite the increasing incidence of pollen allergies, a comprehensive list of allergenic plant species in the Čapljina area has not been compiled to date. This research provides the first systematic analysis of allergenic plants in this area, with a detailed overview of their systematization data, including family affiliation and life form. The paper lists allergenic plant species belonging to three main groups: trees, grasses and weeds. In addition to taxonomic affiliation, plants are also categorized according to their life form, which allows for a better understanding of their botanical significance and potential impact on the aeroallergen spectrum. Although the research does not include an analysis of the seasonality and distribution of these plant species, the collected data represent the basis for future aerobiological research and can serve as a basis for the development of a pollen calendar specific to the Čapljina area. Given the increasing climate change and urbanization, the creation of such lists is important for the early identification of potential sources of pollen allergies and for the adoption of preventive measures that could improve the quality of life of allergic people. The data obtained from this research may be useful for medical professionals, ecologists, pharmacists and public health institutions, enabling a better understanding of local aeroallergens and their potential connection with respiratory diseases. Also, the results may serve as a basis for further research related to air quality, as well as for the planning of urban greenery with the aim of reducing exposure to allergenic plants in populated areas.

Keywords: allergenic plants, pollination, aerobiology, Čapljina

Preparation of biopolymer microparticles filled with vitamin B3 and their antifungal effect - *in vitro*

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ABSTRACT

Biopolymer carriers can improve the stability and controlled delivery of bioactive compounds intended for plant protection. The objective of this study was to prepare vitamin B3-loaded biopolymer microparticles and to evaluate their *in vitro* release behavior and antifungal activity. Microparticles were produced by ionotropic gelation using a natural polymer matrix and calcium crosslinking, followed by encapsulation of vitamin B3. The obtained particles exhibited a stable spherical morphology and a high loading capacity, enabling the gradual release of the active compound in aqueous media. Release kinetics followed the Korsmeyer–Peppas model, indicating Fickian diffusion as the dominant transport mechanism. Antifungal activity was tested *in vitro* against *Botryosphaeria dothidea*, *Fusarium oxysporum*, and *Cryphonectria parasitica*. The prepared formulation inhibited fungal growth by 58.02%, 91.70% and 57.32%, respectively, demonstrating strong biological activity of the released compound. The controlled release from the polymer matrix enabled prolonged antifungal effect compared with non-encapsulated vitamin B3. The encapsulation efficiency of B3 vitamin is 49.33 ± 2.33 %. These results indicate that vitamin B3-loaded biopolymer microparticles represent a promising system for controlled delivery of antifungal agents and may be useful for sustainable plant protection applications.

Keywords: encapsulation, *Botryosphaeria dothidea*, *Fusarium oxysporum*, *Cryphonectria parasitica*, microparticles

Trend analysis of pH values, humus, phosphorus and potassium on agricultural land

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ABSTRACT

Soil fertility monitoring has been conducted continuously since 2003. Since 2019, data have been collected from all authorized laboratories in the Republic of Croatia. A significant increase in the number of requests for agrochemical soil analyses has been observed, indicating a growing awareness of the importance of systematic soil fertility monitoring.

A pronounced upward trend in the number of conducted soil analyses has been recorded since 2019. The peak was reached in 2023, when a total of 39,450 analyses were performed, while by 2025 a total of 33,069 analyses had been recorded. Overall, 193,661 soil samples were entered into the database during the 2019–2025 period.

Trends in soil analysis on agricultural parcels were monitored, and the study included 9 parcels under intensive crop production. On these parcels, agrochemical soil analyses were carried out periodically between 2003 and 2024 at intervals of several years. Long-term changes in key chemical indicators of soil fertility were analyzed, including soil reaction (pH in KCl), humus content, and the availability of phosphorus and potassium.

All analyzed parcels are characterized by predominantly acidic soil reaction. Linear regression analysis did not confirm significant changes in pH values on any parcel ($p \leq 0.005$), indicating a relatively stable but long-term unfavorable soil reaction. Humus content on all parcels falls within the low-humus category. Although changes in humus content were not statistically significant at the strict significance level of $p \leq 0.005$, a consistent downward trend in organic matter content was observed. Phosphorus content showed a very strong negative correlation ($r(\text{trend}) = -0.434$), while statistical analysis reliably confirmed a long-term decreasing trend in potassium content ($p \leq 0.005$).

The results indicate a gradual degradation of soil chemical properties, manifested through pronounced acidity, low humus levels, and a decline in available phosphorus and potassium. Although statistical significance was confirmed only for the decrease in potassium content, the observed trends point to the need for long-term measures aimed at preserving and improving soil fertility.

Keywords: soil, agrochemistry, pH, phosphorus, potassium

Benefits of activating native soil microorganism populations, degrading matured leftovers after harvest

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ABSTRACT

Organic matter (OM) is a key component for the proper functioning of soils. Agricultural practices in recent decades (straw burning, deep plowing, export of straw for energy) have contributed to the depletion of soil OM, leading to a disruption of the soil's native microbial biomass and its biodiversity. On top, climate change complicates the recycling of organic compounds, contributing to the loss of soil productivity. To cope with the current context (input prices and extreme climate), it is crucial to review cultivation practices, which involves restoring the biological mechanisms governing the proper recycling of OM. R&D department of OLMIX SA has developed a non-microbial bio stimulant, intended to develop populations of native microorganisms dedicated to decompose woody crop residues. To support research, two platforms were established in Serbia, led by University of Novi Sad, and in Germany led by the R&D team of an advising company. The platforms were established after the harvest of winter wheat with 5 replicates. Bio stimulant was sprayed on straw; Simultaneously, Levabags were buried at 10 cm. A first soil sample for DNA extraction as control 0 was taken. Four months later, the Levabags were removed from the soil and a second sample for DNA extraction was taken. After their analysis, Leva Lab from Nantes University showed an increase in the amount of straw digested on the bio stimulated plots by 21.8% Vr UNCK. The relative abundance of fungi and bacteria directly or indirectly involved in the processes of straw digestion was profoundly modified. Simultaneously, the relative concentration of pathogenic bacteria was reduced by 17% in Germany and by 58% in Serbia. The relative concentration of pathogenic fungi was reduced by 47% in Germany and by 44% in Serbia. In year 2, the physico-chemical parameters also began to change. Carbon mineralized by the microbial biomass, increased significantly by 17% and nitrate concentration per kg of dry soil was multiplied significantly by 3.6 compared to UNCK. The soil's capacity to absorb water increased by 16%. All of this led to an improvement in yield in Germany by 390 kg ha⁻¹ or 3.8% Vr UNCK, and in Serbia of 880 kg ha⁻¹, or 9.9 % compared to UNCK.

Keywords: soil productivity, Organic matter, Native microbial organisation, soil bio-stimulation

Effects of algae extracts and trace elements on plant bio-stimulation, to activate drought and heat stress resistance pathways

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ABSTRACT

On top of the soil loss in productivity due to non favorable production management use (straw management, tillage, fertilizers...), climate change strongly contribute to the reduction of yield. Excess and important lack of water along the seasons are becoming one of the strongest issue farmers have to face. During drought episodes, even the best irrigation system becomes obsolete; that's why OLMIX SA's R&D innovation team investigated plants drought resistance pathways using the RNA sequencing analysis. RNA-seq analysis highlights the upregulation or downregulation of genes in a plant, allowing us to understand which are the pathways used by plants to respond to stress. In controlled conditions, different crops were cultivated under optimal production conditions. The first group of plants, received four applications of OLMIX biostimulant preventively, while the second, as control group, continued its growth without biostimulant application. Drought conditions were then reproduced and applied to both groups. Once the dry stress established, samples were collected, prepared, and analyzed according to the RNA sequencing method in the research center of DURHAM University. The biostimulant OLMIX under drought conditions has a much greater effect on the plant transcriptome – hundreds of genes changed expression in the biostimulated group. The main pathways affected by the biostimulant OLMIX under drought are related to ion transport, membrane function, cell wall function, and auxin signaling. A field trial was also conducted by Professor Irena JUG from the University of Osijek, in Croatia in 2025, during which the weather conditions were particularly harsh. This trial, carried out according to good experimental practices, showed that soybean plants treated with the Biostimulant OLMIX improved all their yield components, leading them to significantly increase their production.

Keywords: drought, bio stimulant, plant productivity, RNA sequencing

Inhibitory effects of aqueous buckwheat extracts on germination and early growth of redroot pigweed

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ABSTRACT

Buckwheat (*Fagopyrum esculentum* Moench.) is a cultivated species frequently used as a cover crop due to its rapid growth and competitive ability, which contributes to weed suppression. In addition to competition, buckwheat produces secondary metabolites that may affect the germination and early growth of neighboring plants. Redroot pigweed (*Amaranthus retroflexus* L.) is a widespread annual broadleaf weed that causes significant yield losses in row crops. The aim of this study was to evaluate the inhibitory effect of aqueous extracts (10% m/v) prepared from different buckwheat plant parts (whole plant, leaf, stem, fruit) on seed germination and early seedling growth of *A. retroflexus*. The experiment was conducted under controlled conditions (24 °C, 12 h:12 h photoperiod, 70% relative humidity, 40–50 $\mu\text{mol m}^{-2} \text{s}^{-1}$ light intensity). Fifty seeds per treatment were placed in Petri dishes and treated with 4 mL of extract, with distilled water as the control. Germination percentage, radicle length, and hypocotyl length were measured after seven days, and results were expressed as percentage reduction relative to the control. A significant effect of treatment was observed for germination ($p = 0.002077$), radicle length ($p = 2.429 \times 10^{-7}$), and hypocotyl length ($p = 6.961 \times 10^{-8}$). Stem extract reduced germination to 65.5 % and caused the highest reduction in radicle (81.6%) and hypocotyl length (56.1%). Whole plant extract also reduced radicle growth (76.6%), whereas fruit extract showed the weakest inhibitory effect and stimulated hypocotyl elongation. These results indicate that buckwheat stem extracts can influence early growth of *A. retroflexus* under laboratory conditions. However, additional studies are necessary to determine its practical applicability under field conditions.

Keywords: *Amaranthus retroflexus*, cover crops, seed germination, radicle growth, weed

A Multi-Scale Monitoring Framework for Agricultural Water Management: Integrating Cosmic-Ray Neutron Sensing (CRNS) and Satellite Imagery in a Potato Irrigation Experiment

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ABSTRACT

Sustainable agricultural water management requires a comprehensive understanding of soil moisture dynamics and crop responses to irrigation. In this study, satellite-based remote sensing was integrated with field-scale sensing techniques to monitor soil and crop conditions. A field experiment was conducted on potato (*Solanum tuberosum* L.) under three irrigation treatments and a non-irrigated control. A Cosmic-Ray Neutron Sensor (CRNS) was used to provide continuous, non-invasive measurements of average volumetric water content (VWC), representing an effective depth of approximately 35 cm and a spatial footprint with a radius of about 250 m. In parallel, a time-series analysis of Sentinel-1 radar data (VV/VH polarization) was performed to assess temporal variations in surface conditions and their relationship with CRNS-derived soil moisture. A moderate positive correlation was observed between Sentinel-1 VV backscatter and CRNS measurements ($r = 0.61$, $p < 0.05$).

The in-situ experiment demonstrated that non-irrigated control plants were subjected to significantly higher water stress (-14.6 ± 0.43 bar) than irrigated plants (-10.1 ± 0.66 bar), confirming the mitigating effect of irrigation. Due to their relatively large spatial footprints, CRNS and Sentinel-1 observations were not able to distinguish between individual irrigation treatments; however, they provided valuable insights into broader environmental conditions. In particular, CRNS data revealed persistently low background soil moisture during the growing season, underscoring the need for irrigation. Overall, the integration of proximal and remote sensing approaches provides a multi-scale framework for monitoring agroecosystems, supporting precision agriculture applications and contributing to more efficient and sustainable water use.

Keywords: CRNS, soil moisture, water management, precision agriculture, remote sensing

Developing calibration models for the prediction of total carbon content in selected soil types in Croatia using VNIR spectroscopy

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ABSTRACT

The objective of this study was to develop calibration models for predicting soil total carbon (TC) content using diffuse visible and near-infrared (VNIR) spectroscopy for selected soil types in continental Croatia: Stagnosol (Potok), Stagnosol (Hrastovac), Phaeozem (Vukovar), and Luvisol (Lepoglava). Archived soil samples ($N = 99, 35, 72,$ and $17,$ respectively) from depths up to 30 cm were analyzed. Reference TC values were obtained by dry combustion using a CHNS analyzer. Spectral reflectance was measured under laboratory conditions on air-dried, ground, and sieved (<2 mm) samples using a portable spectroradiometer in the 350–1050 nm range, with a sampling interval of 1.4 nm and a spectral resolution of 3 nm. Spectral preprocessing included derivatives, standard normal variate (SNV), multiplicative scatter correction (MSC), and Savitzky-Golay filtering. Partial Least Squares Regression (PLSR) models were developed to predict soil TC content, using only calibration data for Stagnosol (Hrastovac) and Luvisol (Lepoglava) due to limited sample sizes, and a calibration–validation split for Stagnosol (Potok) and Phaeozem (Vukovar). Model performance was evaluated using the coefficient of determination (R^2), root mean square error (RMSE), and the ratio of performance to deviation (RPD). Calibration performance differed between soil types, with RPD_{cal} values of 3.19 for Luvisol (Lepoglava) and 1.09 for Stagnosol (Hrastovac). Validation results showed RPD_{val} values of 3.90 for Stagnosol (Potok) and 1.89 for Phaeozem (Vukovar) in the prediction of TC. These findings support reliable prediction of TC and the use of VNIR spectroscopy as a rapid, cost-effective tool for soil assessment in some Croatian soils.

Keywords: soil total carbon, VNIR spectroscopy, partial least squares regression (PLSR), spectral preprocessing, chemometrics

Encapsulation of *Trichoderma harzianum* T1 in Eggshell Crosslinked Alginate: Characterization and Impact on Pepper Yield and Quality

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ABSTRACT

Sustainable carriers for controlled microbial delivery require environmentally friendly crosslinking sources and stable polymer networks. The objective of this research was to develop calcium–alginate microparticles crosslinked with calcium recovered from eggshell waste and loaded with *Trichoderma harzianum* T1 for controlled release and agricultural application. Eggshell-derived calcium was used to form Ca–alginate microparticles by ionotropic gelation, followed by encapsulation of fungal spores. Fourier transform infrared spectroscopy confirmed calcite-dominant eggshell composition and successful formation of Ca–alginate networks, while spectra of loaded particles showed additional biomolecular bands from the fungal material. Scanning electron microscopy and optical microscopy revealed spherical hydrated particles that shrank after drying, with fungal incorporation increasing porosity from $0.21 \pm 0.09 \mu\text{m}$ to $1.62 \pm 0.55 \mu\text{m}$ and modifying surface morphology. Energy-dispersive X-ray analysis confirmed retention of eggshell-derived Ca within the polymer matrix. Encapsulation resulted in a high loading of 6.8×10^6 spores per particle, an efficiency of 86.1%, and a calcium loading of $1.1 \times 10^{-5} \text{ mol dm}^{-3}$. Release experiments in water showed an initial lag phase followed by a two-stage release with zero-order kinetics ($K_0 = 0.40$ for Ca^{2+} and $K_0 = 0.23$ for spores) and Korsmeyer–Peppas exponents indicating Fickian diffusion ($n = 0.46$ for Ca^{2+} and $n = 0.35$ for spores). The swelling degree decreased from 64.44% in ALG/Ca to 44.49% in ALG/Ca with spores, indicating a stronger network structure. A greenhouse experiment with three pepper cultivars showed improved yield and fruit quality after application of alginate–eggshell Ca microparticles with *T. harzianum* T1, accompanied by higher phenolics, flavonoids, antioxidant capacity, and pigment content. These results demonstrate that eggshell-derived calcium can effectively crosslink alginate to produce stable, bioactive microparticles that enable controlled release of Ca^{2+} and *T. harzianum* spores, providing a low-cost, sustainable approach for crop biostimulation and biocontrol.

Keywords: egg shell, encapsulation, *Trichoderma harzianum* T1, biosimulation, *Capsicum annuum* L.

Effect of Soil Moisture Dynamics on Soil Organic Carbon and CO₂ Emissions from Agricultural Production Sites: A Review

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ABSTRACT

Soil moisture plays a critical role in soil organic carbon (SOC) storage, stabilization, and carbon dioxide (CO₂) emissions amid climatic variability and anthropogenic activities. The soil moisture fluctuation regulate organic matter decomposition, microbial activity and root respiration which impacts on carbon cycle processes. This review synthesizes how moisture regimes under rainfed and irrigated soils drive decomposition of SOC and CO₂ fluxes. Findings from recent studies indicate that moderate soil moisture of 60-70% of water holding capacity enhances SOC stocks and CO₂ efflux is at peak due to high microbial activities. Rainfed soils which has both extreme dry (drought) and wet (saturated) soil triggers drastic pulses of CO₂ emissions. The rare microbes which can survive under drought conditions can be utilized (for rhizosphere modification) to enhance sustainable agricultural production and mitigate climate change risks. This study therefore, confirms that CO₂ efflux and SOC stabilization primarily relies on soil moisture dynamics. The process is complex and its understanding is important for predicting SOC and CO₂ levels in future amid climate variability.

Keywords: soil organic carbon, soil moisture, CO₂ emissions, rare microbes, drought

Comparison of CFA and UV–Vis Methods for Determining Plant-Available Phosphorus in Soil

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ABSTRACT

The objective of this study was to compare two instrumental analytical techniques, Continuous Flow Analysis (CFA) and UV–Vis spectrophotometry, for the determination of plant-available phosphorus in soil, and to evaluate the influence of soil type and selected physicochemical properties on the suitability of the measurement techniques. A total of 165 soil samples representing nine different soil types from eastern Croatia were analyzed. Basic soil properties were determined, including pH (H₂O and KCl), humus content, cation exchange capacity (CEC), carbonate content, hydrolytic acidity, and clay content. Plant-available phosphorus was extracted using the ammonium lactate (AL) method and quantified by both UV–Vis and CFA techniques. The concentration of available phosphorus ranged from 2.14 to 186.3 mg P₂O₅ 100 g⁻¹ (UV–Vis) and from 3.01 to 170.8 mg P₂O₅ 100 g⁻¹ (CFA), depending on soil type. The lowest mean values were determined in pseudogley soils, while the highest were recorded in humus-rich soils. A very strong positive correlation ($r = 0.9972$) was found between the two measurement techniques, with only minor differences in average values. Weak positive correlations were observed between available phosphorus and soil pH, humus content, and CEC, while no significant correlation was found with clay content. The results indicate that both UV–Vis and CFA methods are suitable for determining plant-available phosphorus in soil. However, the CFA technique offers advantages in terms of automation, standardization, and improved interlaboratory comparability of results.

Keywords: ammonium lactate extraction, soil pH, cation exchange capacity, humus content, soil types

The Brown Marmorated Stink Bug (*Halyomorpha halys* Stål, 1855) as a Potential Biological Vector of Aflatoxigenic Fungi

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ABSTRACT

Climate change is intensifying ecological instability in agricultural ecosystems, altering pest distribution and increasing the risk of fungal contamination and mycotoxin accumulation in major crops such as maize. Among emerging threats, the invasive brown marmorated stink bug *Halyomorpha halys* (Hemiptera: Pentatomidae) has rapidly expanded across Europe. This study investigates the capacity of *H. halys* to disseminate mycotoxigenic fungi, with emphasis on aflatoxin B₁ (AFB₁) and ochratoxin A (OTA) producers. A total of 211 pure fungal cultures were obtained from adult maize ears dwelling insects, of which 202 belonged to the genera *Aspergillus* and *Penicillium*. Molecular identification using *BenA* gene sequencing revealed six *Aspergillus* and six *Penicillium* species. *Aspergillus parasiticus* was the most dominant species and mycotoxin screening by thin-layer chromatography confirmed AFB₁ production in 20 isolates, while no OTA-producing strains were detected. The predominance of aflatoxigenic species on *H. halys* suggests that it may play an underrecognized role in the mycotoxigenic fungi dispersal within maize-based agroecosystems. Given its rapid spread, high polyphagy, and capacity to breach plant tissues, *H. halys* may exacerbate mycotoxin risks. Understanding the ecological interactions between *H. halys* and toxigenic fungi is essential for developing effective mitigation approaches to ensure crop safety and protect public health.

Keywords: aflatoxin B₁, *Aspergillus*, *Halyomorpha halys*, maize, mycotoxin contamination

Salinity-Induced Changes in Photosynthetic Performance and Growth of Common Bean (*Phaseolus vulgaris* L.)

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ABSTRACT

Salt stress is one of the major abiotic constraints limiting crop productivity worldwide. Common bean (*Phaseolus vulgaris* L.) is moderately salt-sensitive, yet detailed physiological characterization of its responses to increasing NaCl concentrations remains limited. This study evaluated the effects of salt stress on photosynthetic performance, spectral indices, and plant growth of common bean grown hydroponically under four NaCl concentrations (0, 50, 100, and 150 mM). After salt application, plants were monitored for eight days with measurements every two days.

Photosynthetic parameters were assessed using chlorophyll fluorescence imaging, including maximum quantum efficiency of photosystem II (Fv/Fm), effective quantum yield of PSII (Fq'/Fm'), and non-photochemical quenching (NPQ). Spectral traits were evaluated by multispectral imaging using the normalized difference vegetation index (NDVI) and chlorophyll index (CHI), while morphological traits, digital biomass (DB) and leaf area (LA) were quantified by 3D multispectral scanning. Data were analysed using a mixed linear model with repeated measurements and Tukey's HSD test.

Salt stress induced clear concentration-dependent responses. Fv/Fm remained above 0.75 across treatments, indicating no severe photoinhibition, whereas Fq'/Fm' decreased significantly with increasing NaCl concentration, suggesting reduced photosynthetic electron transport. NPQ increased significantly in salt-treated plants, indicating enhanced thermal energy dissipation.

Growth was strongly inhibited by salinity. Control plants showed a 4.2-fold increase in DB, while plants at 150 mM NaCl exhibited almost no net growth. LA followed a similar trend with reduced expansion at higher salinity levels. In contrast, NDVI and CHI remained relatively stable, suggesting that chlorophyll content was not substantially degraded during the short experimental period.

Keywords: salt stress, *Phaseolus vulgaris*, chlorophyll fluorescence, multispectral imaging, hydroponics

Session 4

Biodiversity Conservation

The effect of bycatch reduction modification of fishing traps on blue crab catch quality

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ABSTRACT

Improving the sustainability of fisheries involves adapting technologies that support environmental conservation without compromising economic viability. In previous work, we showed that modified entrances of fishing traps reduce the by-catch of freshwater terrapin (*Mauremys rivulata*) and have no effect on *Callinectes sapidus* catch rates. We also found that metal cage traps have higher crab catch rates than cylindrical traps. The aim of this research was to determine whether entrance modifications (CON: control, M1: entrance reduction, M2: M1 with the addition of transparent 6 cm net over the funnel) on three trap designs (Cg: metal cage, C1: cylindrical trap with a single entrance, C2: cylindrical trap with dual entrances) affect the size of captured blue crabs. Weight (W), carapace length (CL) and left chela length (LCL) were measured for 235 crabs caught in the Neretva delta during 13 days in February and July 2025. A two-way Welch-type adjustment ANOVA on 20% trimmed means was conducted with robust post-hoc multiple comparisons using the WRS2 package in R. Trap type, modification and their interaction significantly affected LCL ($p < 0.001$, Q: 21.72, 17.72, 21.81). Trap type and trap modification significantly affected CL and W ($p < 0.001$, Q_{CL} : 19.71, 26.84, Q_W : 23.92, 23.45). The Cg:M1 interaction resulted in the largest LCL ($\Psi' = 2.09$ cm, $p < 0.001$) and CL ($\Psi' = 1.45$, $p < 0.001$). W estimates were highest in M1 (154.77 g above average, $p < 0.001$) and lowest in C1 and C2 ($p < 0.001$, -123 g and -132 g, respectively). CON traps of all types consistently showed lower estimates for all analysed traits. Entrance reduction to a fixed ring of 10 cm diameter maximises the harvest of large and heavy crabs. Entrance reduction to 8 cm diameter with a transparent 6 cm net covering the funnel entrance yields crabs with LCL, CL, and W larger than those caught in traps without entrance modification.

Keywords: biodiversity conservation, non-native species control, catch size, *Callinectes sapidus*

Acknowledgment: This research was co-funded by the following projects: (1) LIFE for *Mauremys* (GA: 101071737 – LIFE21-NAT-HR-LIFE for *Mauremys*) granted by the LIFE Programme of the European Union and co-funded by the Environmental Protection and Energy Efficiency Fund and the Government of the Republic of Croatia Office for Cooperation with NGOs; (2) “Ecosystems under multiple stressors: An integrated approach to studying population dynamics of freshwater species” (EkoStres), Grant no: 1-28-AGR11-581: 2025–2027 funded by NextGenerationEU.

SSR Marker-Based Analysis of Genetic Diversity in Croatian Shallot Accessions

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ABSTRACT

Croatian shallots are culturally and agriculturally important crops, yet they remain understudied at the genetic level. Shallot species in Croatia are morphologically diverse, including *Allium cepa* Aggregatum, *A. × cornutum*, and *A. × proliferum*. While the morphological and biochemical traits of these species have been studied, their genetic relationships remain poorly understood. Our study assessed the genetic diversity and population structure of 36 shallot accessions collected from various Croatian regions (Istria, Kvarner, Dalmatia, and northern Croatia). Using 8 SSR markers, 41 alleles were identified, with an average of 5.63 alleles per locus, highlighting substantial genetic variation. Neighbor-net phylogenetic network, STRUCTURE and BAPS analyses revealed distinct genetic groupings, notably separating *A. × cornutum* from other groups. The highest diversity was observed within the *Allium cepa* Aggregatum group. AMOVA results indicated that the majority of genetic variation (57.56%) existed within species, yet substantial differentiation was observed among species ($\phi = 0.42$, $p < 0.0001$). The study provides new insights into the genetic variability of Croatian shallots, highlighting their unique genetic heritage and contributing to the sustainable management of *Allium* germplasm in the region.

Keywords: shallot, genetic diversity, SSR markers, population structure

Biomass of Different Microalgae as a Substrate for Anaerobic Digestion: A Comparative Study

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ABSTRACT

Global increase of fuel demand and its price, increases interests for alternative biofuel production. It is well known that microalgae can be used as biofuel substrate due to their high biomass yield that can be rich in various valuable compounds. However, different biofuel production pathways and the use of different strains, still needs to be researched. This provides a vast platform for different approaches, both in production techniques or in strain selection. Nevertheless, combining microalgae with anaerobic digestion can produce biofuel that represents a widely applicable alternative fuel source. The aim of this study was to compare the biogas potential of different microalgae biomass; chlorella monoculture, cyanobacterial consortia and natural communities. *Chlorella* biomass was used in two different concentrations (5 and 10g). The process of anaerobic digestion was carried out using cow manure in codigestion with the microalgal and cyanobacterial biomass in thermophilic conditions (55 °C) for period of 21 days. The highest biogas yield of 421.40 mL/g volatile solids (VS) (63.97% methane) was achieved using biomass from natural communities. Cyanobacterial consortia had slightly lower yield of 383.34 mL/g VS (64.06% methane). Different concentrations of *Chlorella* biomass showed higher biogas yield of 289.12 mL/g VS (64.25% methane) by adding 10 g, compared to 213.14 mL/g VS (54.78% methane) with 5 g of its dry mass. Based on the obtained results natural communities have potential of higher biogas yield compared to laboratory grown cultures. These results demonstrate the potential of natural communities for biogas production as a sustainable local source of biomass. Also, results highlight how different concentration of microalgal dry biomass in codigestion affect methane yield. In conclusion, utilizing diverse microalgae consortia have a potential for biogas production. However, optimization is needed in order to achieve a commercially viable energy source.

Keywords: algae, biogas potential, cattle manure, thermophilic conditions

Trophic responses of soil nematode communities to *Trichoderma atroviride* bioinoculation in Mediterranean olive orchards

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Abstract

Trophic structure of soil nematode communities is widely utilized to assess ecological responses to management practices in agroecosystems. The aim of this study was to evaluate the effect of bioinoculation with *Trichoderma atroviride* on the trophic structure of soil nematode communities in organically managed Mediterranean olive orchards. A three-year field experiment (2022–2024) was conducted at two locations (Vodnjan and Nadin) on four autochthonous olive cultivars. Two inoculum concentrations of *T. atroviride*: 1×10^6 spores mL⁻¹ (LD) and 1×10^8 spores mL⁻¹ (HD), were applied twice annually. Soil nematodes were identified to genus level and classified into trophic groups. Treatment effects were analysed using generalised linear mixed models (GLMM) with type II Wald χ^2 tests, and cumulative multivariate responses were evaluated using permutational multivariate analysis of variance (PERMANOVA). Bioinoculation significantly altered trophic structure compared with the control. Both doses increased the relative abundance of bacterivores and fungivores and reduced herbivorous nematodes. However, increasing the inoculum concentration did not result in additional improvement of trophic structure. The lower dose produced more balanced shifts, characterised by a stronger suppression of herbivores and a proportionate increase in free-living nematodes. The higher dose did not differ significantly from the lower dose, indicating the absence of additional trophic effects at increased inoculum concentration. PERMANOVA confirmed significant effects of treatment on trophic community structure. The results demonstrate a non-linear, dose-dependent response of soil nematode trophic structure to *T. atroviride* bioinoculation. Optimising inoculum concentration is therefore essential for stable and balanced trophic structure of soil nematode communities in Mediterranean olive orchards.

Keywords: community composition, Mediterranean agroecosystems, microbe–nematode interactions, nematode-based indicators, PERMANOVA, soil health

Has tetrodotoxin become a problem for European seafood consumers?

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ABSTRACT

Tetrodotoxin TTX is a hydrophilic toxin, produced by bacteria that can be found in certain fish species, most commonly the pufferfish, but also marine gastropods and bivalves. It is a potent sodium channel blocker responsible for human intoxication after ingestion of contaminated food. This neurotoxin also poses a food safety challenge due to its heat resistance, making conventional cooking methods ineffective in neutralizing its effects. TTX intoxications were initially mainly associated with human deaths occurring in Southeast Asia, but they have been shown to affect other regions in the Pacific and the Mediterranean as well. There are no health-based guidance values for TTX worldwide, nor maximum levels of TTX in seafood in the European Union (EU), but according to EU legislation, the sale and placing on the market of TTX-containing fish has been prohibited since the last century. European Food Safety Authority (EFSA) has established a safety level of 44 g/kg TTX as the amount of toxin that does not cause adverse effects in humans, but common monitoring has not been performed. Recently, despite the strict ban on its sale and trade, the northern expansion of the yellow-spotted pufferfish, *Lagocephalus sceleratus* (Gmelin 1789), whose northernmost occurrence was confirmed in Croatian Adriatic waters in May 2024, has once again sparked debate about its potential risks to food safety. This invasive species, native to the Indo-Pacific was first recorded in the Mediterranean in 2003. It has now spread to all corners of the basin and is increasingly numerous in the Eastern Mediterranean, where it reaches monstrous sizes compared to the maximum sizes reported in its native area. In this region where the population is established, various measures have been implemented to reduce its population and minimize ecological, economic, and public health impacts. In this review, in addition to the methods used to reduce the population of yellow-spotted pufferfish, possible methods of protecting unaware consumers from accidental consumption are discussed.

Keywords: puffer fish, TTX, food borne diseases, invasive species

Morphological characteristics of traditional plum varieties from the National Plant Gene Bank

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ABSTRACT

In the Experimental Orchard Donja Zelina of the Croatian Agency for Agriculture and Food is maintained the main collection of continental fruit species, which is part of the National Programme for the Conservation and Sustainable Use of Plant Genetic Resources. This paper presents descriptions of 14 plum accessions registered in the Croatian Plant Genetic Resources Database using the CPVO-TP/41/1 plum descriptor. The accessions Bijela kasna mirisava, Brdaklija, Cerićanka, Debeljara, Ilinjača, Kalača, Mandalenka, Motičanka, Pasjara, Pintara, Trnovača and Valpovka originate from Slavonia, while Bistrica and Turkinja are from Lika. Observation and measurement were performed on 44 characteristics of the tree, shoot, flower, fruit, flowering time and ripening time. The fruit weight in all accessions was small, under 30 g, especially in Turkinja, which was under 10 g. The fruit shape was elliptical (Debeljara, Brdaklija, Bijela kasna mirisava and Bistrica), round (Pasjara, Mandalenka, Valpovka and Turkinja), ovate (Trnovača, Cerićanka and Kalača) and obovate (Ilinjača, Pintara and Motičanka). The fruit colour was green (Brdaklija), yellowish green (Cerićanka), yellow (Valpovka), orange-yellow (Pintara, Kalača, Bijela kasna mirisava), dark purple (Ilinjača), violet blue (Pasjara, Mandalenka, Trnovača, Turkinja), and dark blue (Debeljara, Bistrica, Motičanka). Flowering time was early (Cerićanka, Valpovka, Kalača, Pintara, Debeljara), medium (Ilinjača, Pasjara, Mandalenka, Debeljara, Trnovača, Bijela kasna mirisava), and late (Brdaklija, Turkinja, Bistrica). Ripening time was early (Ilinjača, Cerićanka, Valpovka, Pintara, Mandalenka, Turkinja, Motičanka), medium (Pasjara, Debeljara, Kalača, Brdaklija), and late (Bijela kasna mirisava, Bistrica). Analysis of phenotypic classes using the Shannon-Weaver index showed the greatest diversity in fruit colour, flowering time, and fruit ripening time.

Keywords: plum, descriptor, diversity

Performance of a sorting grid in reducing juvenile bycatch in Adriatic Sea bottom trawl fisheries

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ABSTRACT

Bycatch reduction in bottom trawl fisheries can be achieved by improving the size selectivity of the gear. Selective grids are widely used to reduce the bycatch of juvenile fish and crustaceans in bottom trawl fisheries worldwide. However, their performance may vary among fisheries. In this study, we investigated how placing a size-selective grid in front of the codend affects the retention of juvenile bycatch of target species, compared to a standard commercially used bottom trawl in the Adriatic Sea. The two trawls were similar in all aspects except for the final section of the belly, which consisted solely of diamond mesh netting in the standard trawl, whereas in the test trawl it included diamond mesh netting with an inserted size-selective grid. The results showed a significant reduction in *Merluccius merluccius* juveniles in the test gear compared to the standard gear, whereas for other species the findings were either inconclusive or indicated a significant loss of commercially sized individuals. Our results highlight the challenge of simultaneously reducing bycatch and maintaining the retention of commercially valuable catch in multispecies fisheries such as Adriatic bottom trawl fisheries.

Keywords: bottom trawl, bycatch reduction, size selective grid, Mediterranean Sea

Leaf Anatomical Plasticity of Common Bean (*Phaseolus vulgaris* L.) under Water Deficit

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ABSTRACT

Leaf anatomical traits play an important role in plant responses to water deficit. Structural modifications of leaves represent adaptive mechanisms that allow plants to cope with adverse environmental conditions and maintain physiological functioning under stress. The aim of this study was to evaluate anatomical changes in common bean leaves under water deficit. Plants were grown under controlled conditions and subjected to well-watered (control) and water-deficient treatments. After nine days, plants were sampled for structural assessment. Cross-sections were manually prepared from the central part of fresh leaves using a razor blade. Leaf samples were observed using a BM-190 light microscope (BOECO, Germany). Stomatal density was determined from epidermal imprints, while leaf thickness, midrib width, and midrib cross-sectional area were measured from cross-sectional images. All images were analyzed using ImageJ software. Differences between treatments were tested using one-way ANOVA. Significant differences between treatments were observed for all analyzed anatomical traits. Stomatal density increased by approximately 62% under water deficit, which may represent a compensatory response to reduced leaf expansion. Leaf thickness decreased by an average of 17%, likely associated with reduced mesophyll development and loss of cell turgor under drought conditions. Midrib width and midrib cross-sectional area were reduced by approximately 32% and 28%, respectively, indicating a decrease in vascular tissue development and potential limitations in water transport capacity. These results indicate pronounced anatomical plasticity of common bean leaves under water deficit, suggesting structural adjustments that contribute to maintaining leaf functionality under limited water availability. Understanding such anatomical responses may provide valuable insights into mechanisms involved in plant tolerance to water deficit.

Keywords: *Phaseolus vulgaris*, abiotic stress, leaf anatomical traits, structural plasticity

Does biodiversity have two sides?

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ABSTRACT

Preserving natural biodiversity is considered one of the most important challenges faced by ecologists and nature conservationists. However, biodiversity also exists in agricultural production, and this is the biodiversity that we create and that needs to be treated with the same responsibility. Main factors to consider when selecting species in agricultural production should be the quality, safety, and nutritional value they provide to benefit the end user, but the main factors in today's selection of agricultural products are duration of production cycle, disease resistance, production cost, shelf life, transport sustainability, and selling price. Plants from a solanaceous family are perfect examples. The two main food crops with global distribution are potato and tomato. Tomato fruits, aside from being tasty, are healthy as a source of vitamins A and C. Another important nutritional benefit is the lycopene which is a powerful antioxidant that can help prevent many forms of cancer. Unfortunately, the selection of tomato varieties for mass production is based on the ease of cultivation, and suitability for processing, shelf life and suitability for easy transportation, i.e. on profit, and many nutritionally valuable varieties are simply discarded because they do not meet these criteria. Potato strains, or varieties, are classified by shape, color, and processing suitability. Nutritional value and possible health benefits are often forgotten. Historically, primitive forms of cultivated potato and its wild relatives have been used in breeding programs and there is still an enormous potential for discovering desirable characteristics that can minimize the cultivation problem while maintaining nutritional value, safety and quality. Protecting natural biodiversity as scientists, we somewhat forget the biodiversity in agricultural production that we create ourselves. More publicity and scientific research that encourages the development of agricultural crops that bring benefit, health and safety to the consumer should be encouraged.

Keywords: agricultural production, consumers benefit, food safety and quality

Age Structure of Harvested Red Foxes (*Vulpes vulpes* L.) in Nature Park Medvednica – City of Zagreb

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ABSTRACT

The red fox (*Vulpes vulpes* L.), with its extensive geographic distribution encompassing almost the entire Northern Hemisphere, is a highly interesting subject for research. It is notable for its pronounced ecological adaptability, which enables it to successfully inhabit a wide range of habitats. Today, the fox is increasingly present in urban and suburban areas. The aim of this research is to determine the age structure of hunted foxes. Age was estimated by counting dental cementum deposits. Analyses were performed on 255 hunted foxes of known sex over over 12 hunting years (2012/2013 – 2019/2020 and 2021/2022 – 2024/2025). Age was assessed based on cementum annuli in the canines. This research found that the intensity of fox culling shows pronounced annual variation. The analysis did not reveal statistically significant differences in the age of culled foxes. In certain years, an increased proportion of juvenile individuals was recorded, which is associated with higher reproductive success in the population. As fox populations are increasing and are more frequently present in urban and suburban areas, it is important to monitor the abundance and characteristics of the population.

Keywords: red fox, population, J/A ratio, age structure

Satellite monitoring of vegetation dynamics after grassland habitat restoration on Dinara Mountains

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ABSTRACT

During the three-year Dinara back to LIFE project from 2020 to 2023, various grassland habitat restoration techniques were implemented in the DINARA area with the aim of restoring the habitats of three target protected bird species. The area is covered with grasslands belonging to the Eastern Adriatic rocky pastures of the sub-Mediterranean and epi-Mediterranean zones (*Chrysopogono grylli-Koelerion splendidis* Horvatić 1973 and *Saturejion subspicatae* Horvatić 1975) with a successional transition with sharp-needle juniper (*Juniperus oxycedrus*) downy oak (*Quercus pubescens*) and eastern hornbeam, (*Carpinus orientalis*). Overgrown grasslands were restored using three methods: controlled burning, mechanical removal and mixed grazing through the introduction of donkeys as an additional livestock species to existing herds. The aim of this short communication is to present and compare the vegetation dynamics on areas where controlled burning (CB) and mechanical removal (MR) of woody plants and shrubs were carried out during 2021 on two plots, one at 900 m above sea level where controlled burning was carried out (CB-H21) and one in dry fields at the foot of Dinara Mountain at 500 m above sea level (MR-L21). The comparison was carried out by analyzing the trends of the five-year sums of the monthly maxima of the NDVI index derived from the time series of Sentinel 2 images accessed through the Copernicus Browser platform. Results in the period from Jan-2021 to Dec-2025 at both locations CB-H21 and MR-L21 show highly significant positive five-year trends of increasing annual sums of maximum monthly NDVI values, respectively, $y=0.18x+3.7$ ($R^2=0.77$), $y=0.24x+4.7$ ($R^2=0.77$), both $p<0.0003$. It is assumed that the recorded positive trend is a consequence of the secondary growth of cut bushes and rejuvenation. This research demonstrated preliminary results and the potential of using time series of Sentinel-2 satellite images for habitat restoration monitoring.

Keywords: habitat restoration, controlled burning, Dinara, karst grassland, NDVI

Acknowledgement: The results presented in the paper are output from the project „Dinara back to LIFE – Management planning and restoration of Dinara dry grasslands to save biodiversity and support sustainable development“ (LIFE 18 NAT/HR/000847) and Mosaic of LIFE (LIFE24-NAT-HR-Mosaic-of-LIFE/101202651) The projects are co-funded by the Environmental Protection and Energy Efficiency Fund.

The Montado in Portugal: Ecology, Biodiversity and Monitoring in Mediterranean Agroecosystems

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Abstract

The Montado is a Mediterranean silvo-pastoral system of Portugal dominated by cork oak and holm oak. Its open structure results from long-term interaction between human management and natural ecological processes.

This heterogeneous mosaic supports biodiversity, including sclerophyllous vegetation, semi-natural pastures, and fauna structured in trophic networks. The productive base includes Mediterranean shrubs and annual herbs that support herbivores such as the European rabbit, a keystone species. Secondary consumers include insectivorous birds and reptiles, while apex predators such as the Iberian lynx and Spanish imperial eagle indicate ecological integrity. Pollinating and decomposer invertebrates sustain nutrient cycling. The Montado also supports species of European conservation concern, including the cinereous vulture and little bustard, dependent on landscape heterogeneity and connectivity.

This study combined ecological characterization with data from three privately managed agroforestry farms in Portuguese regions, using camera traps, citizen-science records, and georeferenced photographs. Cork oak Montado was the dominant habitat on farms (48.9–59.3%), alongside pastures, eucalyptus stands, wetlands, and scrublands. Despite farm differences, records showed a consistent Mediterranean assemblage of flora, fauna, and fungi. Results show that privately managed Montado can play a crucial role in regional biodiversity conservation when habitat heterogeneity is maintained.

Keywords: agro-silvo-pastoral system, biodiversity, ecological monitoring, habitat heterogeneity, trophic interactions.

Who will save the muddy water fish? Introduction to a conservation project

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ABSTRACT

Most muddy water fish species are considered endangered in Hungary and neighboring countries. Their habitats and populations are threatened by climate change, the introduction of non-native and, in some cases, invasive fish species, as well as ill-considered human activities, such as the dredging of canals – which serve as secondary habitats for these species – using improper methods and at the wrong times. The most important method of species conservation is in-situ conservation, in which organisms are conserved in their natural habitats. Unfortunately, however, in cases where populations are declining too rapidly or the habitats themselves are disappearing, there is no alternative but to carry out ex-situ conservation projects. In Hungary, populations of the European mudminnow (*Umbra krameri*), the Crucian carp (*Carassius carassius*), the Belica (*Leucaspis delineatus*), and the Weatherfish (*Misgurnus fossilis*) are also considered endangered due to the aforementioned factors. For this reason, the National Centre for Biodiversity and Gene Conservation has launched an ex-situ gene conservation program to establish and maintain ex-situ gene banks for these species. This will enable the conservation of these species and the initiation of reintroduction programs. Here, we present our program's main points and initial steps, along with results from our preliminary studies on the genetic variability of Crucian carp, where the results of microsatellite analysis showed that the genetic diversity was moderate (Ho: 0.49-0.61; Ar: 6.01-7.98) in the examined populations. Depending on the genetic structure analysis method, two or three main units with low to moderate differentiation were detected (FST: 0.054-0.192). Based on gene flow, the Danube-Drava region showed a separation from the northern areas and the populations on the eastern bank of the Danube.

Keywords: muddy water fishes, European mudminnow, Weatherfish, Crucian carp, Belica, ex-situ genebank

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***Vkorc1* polymorphism in black rats (*Rattus rattus*) from Adriatic islands: Analysis of exons 1 and 2**

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ABSTRACT

Black rats (*Rattus rattus*) are invasive mammals that threaten seabird populations on several Adriatic islands by preying on eggs and chicks. They are also known carriers of zoonotic diseases that can impact both wildlife and humans. Rodent control commonly relies on anticoagulant rodenticides, which disrupt blood clotting through the VKOR enzyme encoded by the *Vkorc1* gene. As recent studies suggest that variation in exons 1 and 2 may be linked to rodenticide resistance, this study analyses polymorphisms in these regions among black rats from the southern Dalmatian islands. We extracted DNA from muscle tissue and amplified *Vkorc1* exon 1 (n = 30) and exon 2 (n = 57) by PCR, followed by Sanger sequencing. In exon 1, we identified a single polymorphic site at the third position of codon 41, corresponding to a synonymous substitution unlikely to affect warfarin resistance. Among the samples, 12 were homozygous AA, 11 were homozygous GG, and 7 were heterozygous (A/G). Similarly, exon 2 contained one polymorphic site at the first position of codon 94, which was also synonymous (35 homozygous CC, 8 homozygous TT, 14 heterozygous C/T). These results, combined with our previous data on exon 3, confirm the absence of known genetic markers for anticoagulant resistance in the black rat population of the southern Adriatic islands. These findings are crucial for developing effective and sustainable rodent control strategies in this biodiversity-sensitive region.

Keywords: *Rattus rattus*, *Vkorc1*, Exon 1, Exon2, Anticoagulant resistance, Seabird preservation

Impact of *Stereonychus fraxini* on Ash Stand Decline in Plaiul Fagului Reserve, Republic of Moldova

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Abstract

Ash stands represent an important component of forest ecosystems in the Republic of Moldova, providing ecological protection, biodiversity conservation, and valuable timber resources. The objective of this study was to assess the impact of the ash weevil *Stereonychus fraxini* (De Geer, 1775) on the decline of ash stands in the Scientific Reserve “Plaiul Fagului”.

The first outbreaks of the species in the reserve were recorded in 1983 on an area of 1323 ha, with weak defoliation (20–40%) on 78.3% of the affected area, medium defoliation on 16.6%, and severe defoliation reaching 80–100% on 5.1%. During 1983–2025, the extent of outbreaks and defoliation intensity fluctuated under the influence of biotic and climatic factors. Field investigations included preliminary and detailed surveys in stands containing at least 30% ash, as well as pure ash groups with lower participation. Detailed population assessments were based on the density of cocoons in the litter layer, which allowed estimation of the potential defoliation for the following year.

Repeated defoliation caused by adults and larvae reduces photosynthetic capacity, weakens trees physiologically, and increases susceptibility to secondary pests, including other defoliators and xylophagous insects. Current observations indicate localized infestation foci and reduced vitality of ash stands within the reserve. Continuous monitoring and integrated protection measures are necessary to maintain the stability of ash forest ecosystems.

Keywords: ash decline, *Fraxinus*, *Stereonychus fraxini*, defoliation, phytosanitary status

Introducing *Arnica montana* into Hydroponic Cultivation for Sustainable Production

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Abstract

Mountain arnica (*Arnica montana*) is a medicinal plant of high pharmacological value. Arnica flowers (*Arnicae flos*) contain numerous biologically active compounds, including flavonoids, sesquiterpene lactones, essential oils, phenolic acids, and coumarins. Among these, sesquiterpene lactones are particularly important due to their pronounced anti-inflammatory and analgesic effects, which underlie the traditional use of arnica-based preparations in phytotherapy and homeopathy for the topical treatment of contusions, sprains, and inflammatory conditions. In Croatia, *A. montana* is an autochthonous species of montane grasslands, but its populations are threatened by habitat loss, overharvesting, climate change, and abandonment of traditional grassland management, which promotes shrub encroachment and natural succession. Due to harvesting restrictions and increasing demand, arnica is considered a promising crop, although open-field cultivation is limited. The only recognized cultivar, 'Arbo', shows limited adaptability. The aim of ARNIHID project is the introduction of local Croatian populations into hydroponic greenhouse cultivation as a sustainable strategy for the development of production systems capable of meeting market demand for standardized raw material while alleviating pressure on natural habitats. Such efforts could provide a foundation for establishing guidelines for efficient, standardized, and environmentally sustainable arnica production using modern hydroponic techniques for both scientific and commercial purposes. In this context, hydroponic cultivation enables precise control of greenhouse environmental conditions and nutrient solution composition, allowing optimization of plant nutrition and minimization of stress. Consequently, hydroponics represents a particularly suitable cultivation strategy for sensitive and strictly protected plant species such as *Arnica montana*.

Keywords: mountain arnica, *Arnicae flos*, strictly protected species, ARNIHID

Preliminary study of tick infestation in the European wild rabbit (*Oryctolagus cuniculus*) on Goli Otok Island, Croatia

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Abstract

The European wild rabbit (*Oryctolagus cuniculus*) belongs to the order Lagomorpha and the family Leporidae. The species originates from the Iberian Peninsula; however, due to human activities, it is now considered one of the most widely distributed mammalian species in the world. The European wild rabbit inhabits several islands in the central and northern Adriatic region of Croatia. Based on morphological characteristics, it can be concluded that, in addition to wild rabbits, hybrids of wild and domestic rabbits are also present on Goli Otok. The vegetation of Goli Otok is dominated by eastern Adriatic rocky pastures of sub-Mediterranean character and stands of prickly juniper, while a plantation of Aleppo pine is located in the southwestern part of the island. The elevation of the studied locality on Goli Otok ranges from 0 to 100 m above sea level. Goli Otok falls within the humid subtropical climate type with hot summers (Cfa). The mean annual precipitation in this area ranges from 800 to 1100 mm, relative humidity from 60% to 70%, and the mean annual temperature from 13 to 14°C. Ticks were collected from six culled rabbits during regular hunting activities in the autumn of 2025. Ticks were found on all examined individuals. A total of 91 ticks were analyzed, including 34 nymphs and 57 adults. The ticks were attached primarily to the head and neck regions of the rabbits. On average, 15.2 ticks per rabbit were recorded. Tick identification was performed based on morphological characteristics, and all collected specimens were identified as *Ixodes ventalloi*. Based on the obtained results, it can be concluded that the European wild rabbit plays an important role in the population dynamics of *Ixodes ventalloi* on the island, particularly for nymphal and adult stages. However, the role of rabbits in the ecological cycle of certain zoonotic pathogens in this area remains to be investigated, given the known vector potential of this tick species.

Key words: tick, rabbit, island, Goli otok, *Ixodes ventalloi*

Sustainable Cultivation of Marjoram and Basil Using Biostimulants: Evidence from the Nematode Community

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Abstract

Marjoram (*Origanum majorana* L.) and basil (*Ocimum basilicum* L.) are high-value aromatic and medicinal plants that are sensitive to soil stress, including plant-parasitic nematodes. The use of biostimulants in these crops offers a sustainable approach, as they do not act as direct pesticides but instead enhance plant resilience and improve soil biological activity, thereby indirectly reducing nematode-related damage and increasing overall productivity. In this study, three biostimulant treatments were applied to marjoram (M) and basil (B): Bombardier (T1), Rhyzo (T2), and their combination, Bombardier + Rhyzo (T3). The control (C) treatment involved no application of biostimulants. Phytoparasitic genera of nematodes identified in all samples were: *Helicotylenchus*, *Paratylenchus*, *Pratylenchus*, *Rotylenchus*, *Tylenchorhynchus*, *Tylenchus*, and *Xiphinema*. In treatment MT3, six phytoparasitic genera were found, in treatment MT2, only four phytoparasitic genera were found, while other treatments had five phytoparasitic genera each. Genus *Xiphinema* (cp 5) was not found in treatments BT3 and MT2. Treatments MT1 and BT1 had the lowest percentage of phytoparasitic nematodes in the whole nematode community. Notably, treatment BT1 also showed the lowest proportion of highly harmful phytoparasitic nematodes (pp 3–5) in the phytoparasitic community compared to other treatments. The lowest Plant-parasitic Indices (PPI) were recorded in basil treated with Bombardier (BT1), followed by the combined treatment in basil (BT3), and then the marjoram treatments (MT1, MT2, and MT3). These findings indicate that both the crop type and the specific biostimulant applied play a critical role in influencing nematode populations, with basil treated with Bombardier demonstrating the most pronounced suppressive effect on nematode and the highest support for the plant.

Keywords: Bombardier, Rhyzo, phytoparasitic nematodes, phytoparasitic index

Ecological impact and establishment of translocated *Squalius cephalus* in the Lika Region

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ABSTRACT

Squalius cephalus is a widespread cyprinid fish in the Danube basin and one of the most abundant native species in the area. This cyprinid is very resilient and can easily adapt to new environments. In non-native habitats, *S. cephalus* exhibits invasive characteristics such as high fecundity and rapid growth rate. It may cause changes to ecosystems and may prey on native species. The aim of this study was to assess the length–weight relationship (LWR), Fulton’s condition factor (CF), of the *S. cephalus* in the translocated area of the Lika karst region, in comparison with its native range of distribution. Sampling was conducted by electrofishing between July 2021 and July 2022 in the Sava River, which represents the native area of distribution and in three rivers of the Lika karst region representing the introduced range: the Lika, Jadova, and Bogdanica Rivers. The total length and weight of each individual were measured and used to calculate LWR and CF. Data were analysed using Microsoft Excel (2016). A total of 207 individuals were captured in the Sava River, 174 in the Lika, 110 in the Jadova, and 109 in the Bogdanica Rivers. In the native area, isometric growth was recorded in *S. cephalus* individuals from the Sava (LWRb = 3.0521; CF = 1.04442139). In *S. cephalus* from the Lika and Jadova, isometric growth was also observed (LWRb = 3.0579; CF = 0.985183 for Lika; LWRb = 2.9698; CF = 1.00462 for Jadova), while in individuals from Bogdanica (LWRb = 2.8795; CF = 1.056346), negative allometric growth was recorded. Although negative allometric growth was recorded in the Bogdanica River, the CF indicates that individuals are in good condition. However, the results of this study indicate that *S. cephalus* has successfully established stable populations in the Lika karst region and exhibits favorable growth patterns and condition in the translocated area. Hence, *S. cephalus* may have a notable ecological impact on the native fish community, likely through competition for food and habitat.

Keywords: invasiveness, karst rivers, cyprinids, morphology

Acknowledgement: This study was funded by the (1) 'Freshwater fish stock monitoring programme' of the Ministry of Agriculture, Forestry and Fisheries of Croatia and (2) NextGenerationEU project 'Ecosystems under multiple stressors: An integrated approach to studying population dynamics of freshwater species (EkoStres)', Grant no: 1-28-AGR11-581: 2025–2027

GWAS identifies stable QTLs on chromosomes 3A and 4A and highlights the major role of TaMKK3-A in PHS resistance

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Abstract

Pre-harvest sprouting (PHS) is a complex trait in bread wheat controlled by multiple loci involved in seed dormancy and its hormonal regulation. The aim of this study was to dissect the genetic architecture of PHS resistance in a diverse panel of winter wheat genotypes adapted to south-eastern Europe using multi-environment phenotyping and genome-wide association analysis. A panel of 200 cultivars and elite breeding lines was evaluated in four environments using four germination-based assays: whole grain in water (WH), intact spikes (IS), embryos in water (EH), and embryos in abscisic acid (EA), at harvest maturity and 10 days after harvest. Germination index (GI) was used as the main phenotypic trait due to its high discriminatory power and broad-sense heritability. Genotyping was performed using the Illumina Infinium 25K wheat SNP array, and GWAS was conducted with MLM and FarmCPU models on 18,604 SNP markers. A total of 23 QTLs associated with PHS resistance were identified. Although the EA assay showed the highest heritability, the EH assay detected the highest number of QTLs, while chromosome 4A harboured the highest number of associated regions. Two major and stable QTL regions on chromosome 4A (603.3–603.5 Mb and 604.1–604.2 Mb) were consistently detected across assays, environments and models, explaining up to 14.16% of phenotypic variation. Additional stable QTLs were identified on chromosomes 3A, 3D and 5B. The strongest associations were detected in the Phs-A1 region, indicating a major role of TaMKK3-A-linked variation in PHS resistance. These results confirm chromosome 4A as the key genomic region and provide robust markers for marker-assisted selection in wheat breeding.

Keywords: PHS, GWAS, QTL, seed dormancy, TaMKK3-A, chromosome 4A

The effect of fishing trap entrance on the morphometry of turtle bycatch

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ABSTRACT

Non-selective fishing traps pose a life-threatening risk to lung-breathing protected bycatch species. Reducing the entrance size of fishing traps has been shown to decrease the catch rate of chelonian bycatch. Sexual dimorphism in freshwater turtles is generally female-biased in size, with shell shape morphometry adapted for egg production, and fecundity often higher in older females. This research aimed to determine the effect of trap design on *Mauremys rivulata* bycatch morphometry and demographic factors. Two cylindrical trap types (single and double-entrance) were tested with three entrance modifications: control traps (30 cm entrance diameter, 32 mm mesh size), entrance reduction using an 8 cm diameter ring (Mod 1), and Mod 1 combined with a transparent 6 cm net covering the funnel entrance (Mod 2). A total of 141 terrapins were measured over 1,880 soak hours using 40 fishing traps across six days from March to October in the Majkovi Ponds Herpetological Reserve. Two-way robust ANOVA was conducted on maximum carapace width (CW) using 10% trimmed means. Trap modification affected CW ($p < 0.001$, $Q = 24.75$), while no significant main effect was found for trap type or interaction. Mod1 ($\Psi' = 39$ mm, $p < 0.001$) and Mod2 ($\Psi' = 28$ mm, $p < 0.001$) captured significantly smaller turtles than the control group (trimmed CON CL mean = 75 mm, Winsorized variance = 275) in the 10% trim post-hoc comparisons. No significant difference was found between Mod1 and Mod2. A chi-square test with Monte Carlo p-value simulation of the sex-age distribution across trap modifications showed a significant reduction in females in modified traps ($p \leq 0.05$). These results indicate that modifications to fishing trap entrances can reduce adult and female chelonian bycatch in aquatic fauna research, non-native species control, and commercial and recreational fishing.

Keywords: biodiversity conservation, freshwater, *Mauremys rivulata*, fishing trap modifications, demographic factors

Acknowledgment: This research was co-funded by the following projects: (1) LIFE for *Mauremys* (GA: 101071737 – LIFE21-NAT-HR-LIFE for *Mauremys*) granted by the LIFE Programme of the European Union and co-funded by the Environmental Protection and Energy Efficiency Fund and the Government of the Republic of Croatia Office for Cooperation with NGOs; (2) “Ecosystems under multiple stressors: An integrated approach to studying population dynamics of freshwater species” (EkoStres), Grant no: 1-28-AGR11-581: 2025–2027 funded by NextGenerationEU.

An Integrated Morphological, Genetic, and Biochemical Approach to the Identification and Characterization of Wild Edible Mushrooms in Croatia

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Abstract

Wild edible mushrooms represent biological resources of considerable nutritional, ecological, and socioeconomic significance. In Croatia, mushroom species identification remains largely dependent on morphological practices inherently limited by phenotypic plasticity, cryptic speciation, and environmental variability, introducing substantial risk of misidentification with direct consequences for food safety and biodiversity assessments. The present study proposes a structured, multidisciplinary methodology for accurate identification and comprehensive characterization of wild edible mushroom species collected across diverse Croatian habitats. Field sampling includes systematic GPS-referenced collection incorporating seasonal variation and habitat stratification. Morphological examinations, such as macroscopic and microscopic features, will be recorded, while molecular identification, i.e., DNA extraction, ITS barcoding, and phylogenetic analysis will be used for species-level taxonomic verification. Biochemical profiling will be done to quantify nutritional constituents, bioactive compounds, antioxidant activity, enzyme profiles, and fatty acid composition. Microelements and potentially toxic heavy metals will be determined in mushroom tissue and corresponding soil samples. Food safety evaluation will be done using bioaccumulation factors, health risk indices, dietary intake modelling, and target hazard quotients. The proposed workflow establishes standardized, reproducible protocols applicable to understudied regional ecosystems. Thus, integration of molecular taxonomy, biochemical profiling, and environmental analysis strengthens food safety monitoring, supports evidence based biodiversity conservation, and enables sustainable utilization of wild edible mushroom resources.

Keywords: wild edible mushrooms, ITS barcoding, biochemical profiling, heavy metals, food safety

Acknowledgement: This paper was supported by the Croatian Science Foundation (HRZZ) under the project Integrated morphological, genetic, and biochemical identification and characterization of wild edible mushrooms in Croatia - GenBioM (IP-2025-02-9719).

Shell shape variation of thick-shelled river mussel (*Unio crassus* complex) in Croatia

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ABSTRACT

The thick shelled river mussel (*Unio crassus*) is an endangered European freshwater bivalve. It inhabits a wide variety of streams and rivers in continental Croatia. Recent studies indicate that *U. crassus* is a species complex comprising several distinct taxa at both the European and Croatian levels. Its shell morphology exhibits a high level of environmental plasticity, resulting in diverse shell forms across different habitats. Due to a lack of comprehensive data on this complex in Croatia, the aim of this research was to determine the shape variation of *U. crassus* across diverse habitats within the Danube basin in Croatia. Individuals were collected in autumn 2025 from 18 sites across nine rivers, with a minimum of 11 individuals per site used for geometric morphometry analysis. Individuals were photographed, and the obtained images were digitized by 42 landmarks. Data were tested for digitization error, and the dimensionality of the data was reduced via principal component analysis. The allometric effect was removed prior to a permutational multivariate analysis of variance. Discriminant analysis was employed to visualize group separation, while shape changes were illustrated using wireframe graphs. Results indicate significant shell shape variation among several rivers, whereas intra-river variation was generally non-significant, with the exception of the Ilova River. Sites on the Kupa River, as well as specific sites on the Korana and Ilova rivers, showed the greatest morphological divergence. Shape variation of this complex may result from phenotypic plasticity, taxonomic heterogeneity, or both, where similar microhabitats may drive convergent morphologies. In the absence of population genetic data, further research is required to fully resolve the morphological and taxonomic composition of this complex in Croatia.

Keywords: geometric morphometry, freshwater bivalves, lotic habitats, Danube basin

Acknowledgement: This research was co-funded by the following projects: (1) “Ecosystems under multiple stressors: An integrated approach to studying population dynamics of freshwater species” (EkoStres), Grant no: 1-28-AGR11-581: 2025–2027 funded by NextGenerationEU; (2) 'Multifaceted approach to understand freshwater mussel decline and resilience in the face of global warming - FACEMUSSEL' funded by the Swiss National Science Foundation - SNSF (MAPS - No. IZ11Z0_230666) and Croatian Science Foundation - HRZZ (MAPS-2024-9713).

Preliminary assessment of the effectiveness of commercial entomopathogenic nematodes in controlling the *Ceratitis capitata* under laboratory conditions

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Abstract

The Mediterranean fruit fly, *Ceratitis capitata* Wiedemann, 1824 (Diptera: Tephritidae), is one of the economically important pests of fruit crops worldwide. Recent research has confirmed the ability of the pupa to overwinter in the soil as one of the overwintering stages, and the conditions in the Neretva Valley with the availability of hosts such as peach, fig and mandarin are ideal for the development and maintenance of the population in that area. Entomopathogenic nematodes are an effective method of biological control of the Mediterranean fruit fly, which could easily be integrated into the existing integrated plant protection program in the Neretva Valley. The aim of this work is to preliminarily determine the effectiveness of the commercially available nematode *Steinernema feltiae* in controlling *C. capitata* under laboratory conditions. *C. capitata* pupae were collected from infected mandarin fruits (*Citrus reticulata*) in the Neretva Valley in the autumn of 2025, as well as soil samples used in the study. The experiment was carried out in three repetitions, at room temperature for 99 days, during which the mortality of the treated pupae was monitored daily, that is, the development of the insect to the adult stage and the life span. Twenty pupae were placed in pots (1000 mL) with soil, after which the soil was treated with entomopathogenic nematodes (infective larvae, IL). Two doses of entomopathogenic nematodes diluted in water were used, a higher dose containing 15000 IL in 10 mL and a lower dose of 7500 IL in 5 mL, as well as a control (untreated samples). After the mortality of all adults emerging from the treated samples, the mortality of untreated individuals was read every 5 days. At a higher dose of nematodes, mortality of 96-100% was determined within 4-6 days, while at a lower dose, mortality of 85-92% was determined within 5-7 days. In the control, mortality was 25-35% within 5-7 days, and 50% between 15-40 days, depending on the date of the experiment. In monitoring the emergence of adults, mortality was recorded at the higher dose of 50%, the lower 35-40%, and the control of 15-20%. Both nematode treatments showed a statistically significant difference in mortality and pest development compared to the control treatment. These preliminary research results indicate the effectiveness of entomopathogenic nematodes in controlling the pupa of *C. capitata*, which could be applied selectively on individual trees of wild and cultivated host plants.

Keywords: *Ceratitis capitata*, *Steinernema feltiae*, mortalitet

Genotype × year interaction and yield stability of F6 pea breeding lines

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Abstract

This research aimed to assess yield stability in the F6 generation of fodder pea breeding lines through four growing seasons and to determine the contribution of yield components to the total yield. Nine F6 genotypes were analyzed during four growing seasons in a randomized block design experiment with three replications. Grain yield and yield components were analyzed: number of pods per plant, number of grains per pod, and weight of 1000 grains. The G × Y interaction was analyzed using the additive main effects and multiplicative interaction model (AMMI) and Finlay–Wilkinson regression. The results showed a significant interaction, indicating that the genotypes responded differently across years. AMMI analysis showed that the first principal component of the interaction explained most of the variability, whereas the stability parameters enabled discrimination of genotypes based on their adaptability. Among the analyzed genotypes, genotype 3 showed the most favorable combination of yield and stability, with an average yield above the trial average and a uniform yield throughout the years. In contrast, genotype 9 achieved the highest yield, but with low stability, while genotype 7 showed the highest stability, but with the lowest yield. Regression analysis indicated the presence of genotypes with broad and specific adaptability. Path analysis showed that the number of pods per plant has the largest direct effect on yield, followed by 1000-grain weight, while the number of grains per pod has a smaller contribution to yield formation. The obtained results indicate that the stability of yield in the analyzed material is primarily determined by the interaction G × Y, while the number of pods represents a key component for yield improvement in the early generations of the breeding process.

Keywords: Genotype x environment interaction, yield stability, yield components, *Pisum sativum* L.

Acknowledgments: This paper is the result of research within the internal competitive project of the Agricultural Institute Osijek, "Genotype selection of peas in the framework of climate changes" (2024. – 2027.), which is funded by the European Union—Next Generation EU through the National Recovery and Resilience Plan 2021. – 2026.

Morphological characteristics of five authentic olive varieties in Zadar County monitored within the Central BIC project

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ABSTRACT

The Zadar County area has a centuries-old tradition of olive cultivation, where the variety represents the basis for the production of authentic olive oils, due to its significant influence on the composition and quality of the fruit. Zadar County, as a partner, is participating in the implementation of the CENTRAL - BIC – Central Europe Biodiversity Innovative Communities project as part of the Interreg CENTRAL EUROPE 2021–2027 Programme. In line with the objectives of the project, five indigenous olive varieties were selected ('Drobnica' – Karin, 'Plavica' – Pakoštane, 'Peljužica' – Sukošan, 'Peljužica' – Pašman and 'Masnjača' – Lovinac) for morphological analysis of the fruit. The analysis included measuring the mass of the fruit and the stone in order to calculate the mass and content of the mesocarp. The length and width of the fruit and stone were also measured according to the international COI method. Mean values were compared using one-way analysis of variance (ANOVA), and significant differences were determined using the Tukey post hoc test using Statistica 13 program. The results showed differences between the varieties, with the 'Plavica' variety distinguished by the largest fruit and the highest proportion of mesocarp. According to the classification of fruit shape based on the ratio of the length to the width of the stone, all varieties have an elliptical fruit shape (L/W 1.8 – 2.2) except for 'Masnjača' which has an oval fruit (L/W 1.4 – 1.8). As morphological characteristics, in addition to genotype, are influenced by growing conditions, a reliable assessment requires long-term monitoring of varieties under uniform agro-ecological conditions.

Keywords: biological diversity, fruit shape, 'Masnjača', mesocarp content, 'Plavica'

Session 5

Agribusiness and Smart Agriculture

Relationship between spray coverage and liquid spray deposition as indicators of spraying process quality

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ABSTRACT

The study investigates the relationship between spray coverage and liquid spray deposition as indicators of spraying process quality under real field conditions. Field experiments were conducted using a field sprayer under varying operational parameters, including nozzle type (standard and air induction), pressure (200 and 400 kPa), and driving speed (5 and 10 km·h⁻¹). The research was conducted using a Unia Pilmet Plus 2521 sprayer (Poland). Spray coverage was assessed using water-sensitive paper attached to artificial plants, while liquid spray deposition was determined based on boron content in the plant material. Statistical analyses, including Pearson correlation and linear regression, were applied to evaluate the relationship between these parameters. The results demonstrated a strong positive linear relationship between spray coverage and boron content, with coefficients of determination (R^2) exceeding 0.85 in most cases. The weakest relationship was observed for the XR nozzle at low pressure (200 kPa) and low speed (5 km·h⁻¹). Higher sprayer speeds generally resulted in improved model fitting. These findings indicate that spray coverage can serve as a reliable and practical indicator for estimating liquid deposition. The proposed approach provides a simple and effective tool for assessing spray application quality in agricultural practice.

Keywords: spray coverage, liquid spray deposition, nozzle, sprayer

Launching an operational group dedicated to climate-friendly olive growing

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ABSTRACT

Strengthening the synergy between farmers and researchers is crucial for solving practical agricultural challenges. To support this goal, the Common Agricultural Policy Strategic Plan implements Intervention 77.03 – Operational Groups within the European Innovation Partnership for Agricultural Productivity and Sustainability (EIP-AGRI), a framework dedicated to improving agricultural productivity and sustainability through collaborative innovation.

This abstract presents preliminary findings from an EIP-AGRI operational group project that integrates primary and secondary data to propose climate-friendly management measures for olive groves in the western part of the Istrian Peninsula, Croatia. Data were collected using a three-step method. First, climatological data from the study area were analysed over a six-year period. Second, potential climate-friendly management practices suitable for the identified climatic conditions were evaluated. Third, primary data were gathered through interviews and focus group discussions with olive growers participating in the project.

The results reveal a sensitive balance between temperature and precipitation, emphasising the need to implement climate-friendly practices to ensure yield stability and maintain high-quality olive oil production. Analysis of primary data offers insights into the current conditions of the olive groves included in the project and the farmers' knowledge of climate-friendly practices. Additionally, the study presents the management practices selected by the farmers for implementation during a two-year field research period.

Keywords: EIP-AGRI, climate change, olive cultivation, climate-friendly practices, three-step method

Agroforestry business models for climate-resilient production

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ABSTRACT

Climate change (CC) and other risks, such as price volatility and the geopolitical situation, affect agricultural production. The Pannonian Biogeographical Area (PBA) has the second largest share of agricultural land use in Europe and ranks first in productivity. In recent years, the PBA has also faced extreme weather conditions, which are being researched within ClimaPannonia, an EU-funded Horizon project. The objective of this research is to present agroforestry practices as a sustainable business model (BM), examining key production and market challenges in berry production. Data describing the current situation and supporting the proposed agroforestry BM were gathered during a workshop in Fertőd, Hungary. The workshop brought together researchers, students, berry producers, and non-governmental organisations active in agroforestry and sustainable agriculture. Berry production was chosen because it is the most important type of production in this region of Hungary. The main results show that participants identified CC as the primary factor affecting agricultural production, as seen in decreased yields and changes in fruit quality due to heat stress, drought, and shortened ripening periods. Additionally, labour shortages and saturated fresh-berry markets were identified as major risks. Therefore, innovative BM such as organic production, differentiation through processing, providing premium quality, cooperation among farmers, and improved use of ecosystem services, carbon-related opportunities, circular economy, and offering diversified products or services may be considered for climate-resilient production.

Keywords: agroforestry, business model, climate change, innovation, sustainability

Cluster analysis of agricultural producers in the Posavina LAG area

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ABSTRACT

Empirical research was conducted in the area of LAG Posavina on a sample of 328 agricultural holdings (AH) with the aim of identifying homogeneous groups of producers based on the development characteristics of their AH, perception of obstacles, and degree of cooperative orientation. Data were collected using a questionnaire, and both descriptive and inferential statistical methods were applied in the analysis. For segmentation, K-means cluster analysis was used, with variables covering structural, market, institutional and entrepreneurial development factors included in the model.

The results identified three statistically differentiated clusters. The first cluster (55%) consists of moderately proactive individualists, characterised by a relatively higher level of market activity but limited willingness to engage in more intensive cooperative networking. The second cluster (38%) includes passive and unconnected producers with a lower level of development initiative, weaker involvement in projects, and a more pronounced perception of market constraints. The third cluster (7%) consists of formally involved but developmentally disengaged entities, who participate in certain forms of association but lack significant strategic orientation.

Analysis of variance confirmed statistically significant differences between clusters in the perception of market and entrepreneurial barriers, level of education, participation in development programmes, and degree of cooperative orientation. Producers with a stronger propensity for association demonstrate greater development dynamics and a higher level of market integration. The results confirm the existence of heterogeneity within the observed sector and indicate the need for a differentiated approach in shaping development policies and support measures.

Keywords: agricultural holdings, cluster analysis, development factors, development policies

Impact of current CAP subsidies on farm income level and stability in Croatia

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ABSTRACT

In 2024, approximately €1.02 billion in subsidies was paid to farmers in Croatia from two main Common Agricultural Policy (CAP) funds: EAGF and EAFRD. This amount includes both current subsidies and investment grants. This research examines the extent to which total current CAP subsidies, excluding investment support, influenced the level and stability of income for Croatian agricultural holdings during 2014–2023. The analysis is based on individual panel data from the Croatian Farm Accountancy Data Network (FADN), covering 1,290–1,408 holdings annually. In the first stage of analysis, descriptive statistics were used; to determine causal effects, a fixed-effects panel regression model and a cross-sectional regression model were applied. To measure income volatility, the standard deviation of the inverse hyperbolic sine-transformed gross income (SE410) and net income (SE420) was used. Monetary variables for the period 2014–2022 were in HRK, and the fixed exchange rate of the Croatian kuna to the euro was used to convert values to EUR (1 EUR = 7.53450 HRK). All monetary variables were deflated using the GDP deflator to obtain real values.

The results of the fixed-effects model show that changes in the absolute amount of subsidies (SE605) within the same holding do not have a statistically significant impact on either gross or net income. In contrast, the intensity of subsidies, measured as the ratio of total subsidies to total output (SE131), is negatively associated with both gross and net income, with a statistically significant effect for net income. Economic size (SE005) has a positive and robust effect on income level in all specifications. Cross-sectional results indicate that average subsidy intensity does not significantly affect gross income volatility, but it is positively associated with net income volatility. Significant differences in income volatility are observed across production types, economic size classes, and regions, with higher volatility in Continental Croatia.

The findings suggest that CAP subsidies in Croatia primarily play a redistributive role rather than directly increasing income. Greater relative dependence on subsidies is associated with lower income levels and, for net income, higher volatility, indicating structural weaknesses in more subsidy-dependent holdings.

Keywords: CAP, farm income, income volatility, subsidy intensity, Croatia

Digital agriculture awareness and knowledge among family farm holders in Osijek-Baranja County

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ABSTRACT

The study aims to examine the level of awareness and knowledge of digital agriculture concepts and technologies among family farm holders in Osijek-Baranja County. Data were collected through online and face-to-face surveys conducted in the summer of 2022 on a sample of 43 family farmers. The questionnaire investigated farmers' familiarity with selected terms related to digital agriculture, the presence of potential barriers for implementing digital technologies, and the use of digital tools in farm management and marketing. Respondents were also asked about their use of websites, social media promotion, and experience with European Union funding schemes. Additionally, farmers evaluated their agreement with statements describing possible constraints on the adoption of digital solutions in agriculture. Most respondents were male (76.7%) with an average age of 42 years. For more than half of the participants (56%), agriculture is the primary source of income, while 58% reported having higher education. A large proportion of respondents had been involved in agricultural activities for less than ten years (44%), and the majority operated farms with only one employee (69%). Participants represented diverse agricultural sectors, including rural tourism, beekeeping, crop production, organic farming, livestock production, and viticulture. The results indicate that farmers are generally familiar with key digital agriculture concepts; however, practical application remains limited. Almost half of the respondents (45.5%) reported not using ICT tools in their daily business operations, and approximately two thirds of farms do not maintain an official website. In contrast, social media platforms are widely used, and farmers expressed moderate satisfaction with the availability of internet infrastructure. Identified barriers to the adoption of ICT solutions – such as limited time, insufficient knowledge, and lack of labour – were found to be associated with farm characteristics, including the type of agricultural production, income category, VAT system participation, and the main source of household income. The findings suggest that the digital transformation of Croatian agriculture still offers significant potential. Strengthening advisory services and educational programmes could play an important role in facilitating the wider adoption of digital technologies and supporting the transition towards more digitally enabled farming systems.

Keywords: family farms, digital agriculture, ICT adoption, farmers' knowledge, education

University students' acceptance of Mediterranean-type foods in a canteen setting and its determinants: Evidence from Zagreb, Croatia

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ABSTRACT

Public health and institutional food systems are increasingly emphasising the promotion of healthy eating habits among university and college students. This study examines students' acceptance of newly introduced Mediterranean-style foods in a university canteen and identifies the main factors influencing their choices. The analysis evaluates the impact of socio-demographic characteristics, living arrangements, eating habits, and institutional factors such as menu satisfaction, pricing, and promotional activities, using data from 744 surveys and applying descriptive statistics and a multivariate probit model. Students who actively engaged with promotional materials were significantly more likely to select Mediterranean options, indicating that knowledge and information play a crucial role in shaping food choices. Convenience and health perception emerged as the primary factors for acceptance, while price sensitivity, satisfaction with existing menus, and a preference for familiar foods were key barriers. Notably, there was a discrepancy between students' expressed desire for a more varied menu and their actual purchasing habits. To promote healthier food choices, these findings highlight the importance of combining menu innovation with effective communication strategies and competitive pricing. The results provide practical insights for developing targeted interventions to encourage the adoption of Mediterranean-style diets in institutional food environments, relevant for canteen managers, policymakers, medical professionals, and other stakeholders in institutional food provision.

Keywords: mediterranean diet, university students, food choice behaviour, institutional food environment, multivariate probit model

Evaluation of the effectiveness of public support in multifunctional rural development

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ABSTRACT

Measuring the effectiveness of public support is essential for policymakers, researchers, and stakeholders to assess the impact of interventions and allocate resources efficiently. However, there is a lack of empirical studies that systematically evaluate these effects in Croatia. The four-year EVA-MURA project addresses this gap by assessing how public support influences multifunctional rural development in Croatia. Its aim is to analyse economic diversification, social well-being, and environmental sustainability, and to provide actionable recommendations for improving future rural policies. The project combines the quasi-experimental method Propensity Score Matching with field research and participatory approaches to identify the actual effects of support measures. A central innovation is the Rural Multifunctionality Index, a composite indicator that quantifies a wide range of support effects, including the development of supplementary and non-agricultural activities, landscape preservation, local value creation, and social innovation at the county level. This index enables comparison across counties, progress monitoring, and more targeted allocation of public funds. EVA-MURA advances evaluation methodology, develops a digital database, and offers concrete proposals for enhancing multifunctionality, supporting evidence-based rural development aligned with EU long-term rural visions, and strengthening research capacity and open science.

Keywords: multifunctional rural development, Propensity Score Matching (PSM), effectiveness, support measures, Rural Development Multifunctionality Index

Typology of family dairy farms in Pannonian Croatia: Evidence from cluster analysis

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ABSTRACT

Pannonian Croatia, the largest NUTS 2 region in the Republic of Croatia, is located in the eastern part of the country and represents the core area of national agricultural production as well as one of the main centres of the Croatian dairy sector. This research analyses the structural and production characteristics of dairy farms in this region, aiming to identify typological groups based on similarities in resources and production indicators. The analysis covers all 1,584 family dairy farms in 2022 and includes the number of dairy cows, number of crops, utilised agricultural area (UAA), and milk yield per cow as an indicator of productivity.

Cluster analysis identified 16 farm types. Clusters 1–3 include farms with 8–14 cows and account for 69% of all farms observed. These farms have lower milk yields (3,678–3,950 kg per cow), indicating predominantly small, less intensive, and partly mixed operations. Their production is below the national average (4,210 kg), suggesting limited resources and lower specialisation. In contrast, Clusters 4–6 represent large and more intensive farms (49 to 212 cows) with above-average production (5,421–8,061 kg per cow) and smaller area per head, indicating strong concentration and efficient use of resources. The two largest farms, with 456 and 317 cows from separate clusters, reflect their structural specificity and high production concentration.

Overall, the sector is characterised by a strong dominance of small farms, while intensive production is concentrated in a small number of large farms, reflecting trends observed elsewhere in the EU. The resulting typology improves understanding of the sector's structure and provides a basis for targeted agricultural and rural development policies, particularly given the role of these farms in maintaining agricultural land in use.

Keywords: family dairy farms, cluster analysis, Pannonian Croatia, farm typology

Motives and factors of consumer's choice between natural and artificial Christmas trees in Croatia

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ABSTRACT

The choice between natural and artificial Christmas trees is a complex consumer decision influenced by demographic, economic, and perceptual factors. The aim of this study was to identify the main reasons influencing consumers' decisions when selecting a type of Christmas tree, and to examine the relationship between socio-demographic variables and consumer preferences. The research was conducted using a structured online questionnaire with a sample of 1,050 respondents. Descriptive and comparative statistical analyses were used to evaluate differences between users of natural and artificial Christmas trees.

Results show that artificial Christmas trees were used by 57.5% of respondents, while 42.5% preferred natural trees. People living in urban areas were more likely to use artificial trees, whereas respondents from rural areas showed a stronger preference for natural trees. Practicality (84.11%) and cost-effectiveness were the most important motives for choosing artificial trees, while tradition (78.03%), natural scent (77.35%), and the creation of an authentic holiday atmosphere (65.70%) were dominant motives for choosing natural trees. Environmental reasons were present in both groups, but were not the primary factor in decision-making. Respondents also tended to be subjective regarding the environmental justification of their preferred tree type, as most considered their own choice to be more environmentally acceptable. Additionally, users of natural trees showed a 3.5 times higher probability of considering a switch to artificial trees compared to users wishing to change from artificial to natural trees.

The results confirm that consumer choice of Christmas tree type is determined by the interaction of functional, cultural, and socio-economic factors. These findings contribute to a better understanding of consumer behaviour related to seasonal forestry products and provide a solid basis for further research into sustainable consumption and environmental perception in forestry-related markets.

Keywords: Christmas trees, consumer behaviour, artificial trees, natural trees, sustainability

Navigating the high-protein era: Croatian consumer intentions and drivers for dairy protein beverages consumption

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ABSTRACT

Dairy protein beverages have become increasingly popular due to rising consumer interest in health and convenience; however, the factors driving their consumption remain only partially understood. This study investigated the determinants of consumer intention to consume dairy protein beverages in Croatia. An online survey was conducted among 265 Croatian consumers, measuring behaviour during purchase and consumption of dairy protein beverages, food-related lifestyle, health consciousness, attitudes towards dairy protein beverages, subjective norm, and consumption intention, using a 5-point Likert scale (1 – strongly disagree to 5 – strongly agree). Multiple linear regression analysis showed that subjective norm is the strongest predictor of intention to consume dairy protein beverages, explaining 27.1% of the variance, followed by attitudes and health consciousness. Together, these three variables explained 35.9% of the variance in consumption intention, while food-related lifestyle did not significantly contribute. Among consumers, most prefer classic flavours such as chocolate and vanilla, and domestic brands, with the majority of purchases made in-store rather than online. Respondents with a stronger intention to consume dairy protein beverages also consumed them more frequently. These findings suggest that positioning dairy protein beverages as healthy, tasty, and easily available, while leveraging social influence through media and peer recommendations, is likely to enhance consumer intention to consume these beverages.

Keywords: consumer behaviour, dairy protein beverages, intention, subjective norm

Understanding game meat consumption through health and environmental goals: A theory of reasoned goal pursuit framework

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ABSTRACT

One of the key challenges in the coming decades will be to ensure a safe, nutritionally adequate, and sustainable food supply. Given limited natural resources and the environmental pressures associated with intensive livestock production, there is an increasing need to diversify sources of animal protein. Game meat is increasingly recognised as a potentially sustainable alternative to conventionally produced meat. Additionally, it represents a nutritionally valuable food source, while wild game also plays an important role in biodiversity conservation.

The Theory of Reasoned Goal Pursuit (TRGP) extends the Theory of Planned Behaviour (TPB) by incorporating goal-directed processes into the prediction of human behaviour. Building on this theoretical foundation, the present study aims to examine both behavioural determinants and health and environmental perceptions influencing consumer acceptance of game meat.

The main objective of this research is to identify the key factors influencing consumers' intention to consume game meat, using an integrated TPB–TRGP framework. Data will be collected through an online survey conducted on a representative sample. Statistical analyses will be performed using SPSS (version 30) and SmartPLS (version 4).

As the research is currently ongoing, only expected results can be presented at this stage. It is anticipated that integrating TPB and TRGP will provide a more comprehensive explanation of consumer intentions towards game meat compared to the traditional TPB model alone. Attitudes are expected to emerge as the primary determinant of intention, particularly when shaped by health-related beliefs and perceived benefits of game meat consumption. Subjective norms are also expected to play a significant role, especially when consumers are socially connected to hunters. Perceived behavioural control is anticipated to influence intention primarily through perceived availability of game meat.

Furthermore, variables introduced through the TRGP framework – particularly health-related and environmental goals – are expected to play a central role in shaping consumption intentions. To the best of the authors' knowledge, no previous study has applied an integrated TPB–TRGP framework to the context of game meat consumption, nor has it comprehensively explained consumer intentions towards this specific product category.

Keywords: game meat, consumers, questionnaire, intention to eat

Design and implementation of a cow monitoring system

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ABSTRACT

This poster presents a vision-based system for continuous, non-invasive monitoring of dairy cattle in barn conditions, designed to support early detection of health and welfare issues, and discusses problems encountered during the design and implementation of the hardware, such as connectivity issues, data transfer, volume of data, etc. The process was repeated in four barns. Each barn was equipped with a network of 20 RGB cameras, and the acquired data were transferred via Ethernet to a PC for further processing. Monitoring was performed for approximately 18 h per day, between 05:00 and 20:00, over a period of 2 months in each barn. From the collected recordings, 8,000 randomly sampled frames were manually annotated by human annotators. Annotations provide cow-oriented bounding boxes, animal state (standing or lying), head orientation, and whether the cow is fully or partially visible. From these data, we developed deep learning methods for cow detection and tracking as the core of a monitoring pipeline. We trained a YOLO11n-OBB deep learning model for cow detection, using oriented bounding boxes to better represent animal extent and pose in barn scenes. For tracking, detections were linked over time using a standard tracking-by-detection approach based on IoU matching, Kalman filter motion prediction, and Hungarian assignment. The detection component reliably localizes animals in complex barn scenes, and the tracking methods produce continuous trajectories required for longitudinal analysis of individuals across cameras and a basis for future development of deep learning methods for individual cow identification. This process establishes a foundation for automated analysis of individual animals over time and is a necessary step toward behaviour classification and early disease detection (e.g., lameness).

Keywords: cow monitoring, animal welfare, computer vision, deep learning, multi camera system

UAS-Based RGB High-Throughput Phenotyping for Agronomic Trait Extraction in Maize

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ABSTRACT

Unmanned aerial system (UAS)-based RGB imaging has emerged as a cost-effective high-throughput phenotyping approach for rapid, efficient, and non-destructive assessment of important agronomic traits in maize. A diversity panel comprising 200 maize hybrids and their parental components (400 genotypes in total) was evaluated for plant density and height under field conditions. RGB imagery was collected weekly throughout the maize growing season and twice weekly during pollination using a DJI Mavic 3M UAS platform set to a ground sample distance (GSD) of 0.42–0.54 cm pixel⁻¹. The trial was conducted at two locations in 2025 and the resulting RGB orthomosaics and digital elevation models (DEMs) were processed for plot-level data extraction. UAS-based RGB pipelines were developed for high-throughput extraction of plant density and plant height in maize from RGB orthomosaics and DSM, respectively. From early-stage RGB orthomosaics (2–5 leaf stage, prior to canopy overlap), the developed script enables automated detection and counting of individual plants across plots. By integrating RGB-based vegetation segmentation, plot-wise row orientation using principal component analysis (PCA), and spatial clustering, the workflow generates stable plant counts under varying field conditions while reducing noise from soil background and inter-row areas. Plant height is derived from UAS data using a digital elevation model (DEM) differencing approach between vegetation and bare soil surfaces. Together, these scripts enable efficient plot-level phenotyping from RGB imagery, providing a practical basis for large-scale field experiments. For both traits, ground-truth measurements have been collected manually as reference data for validation of traits extracted and calculated from UAS imagery. The proposed framework is expected to support scalable and accurate phenotyping pipelines for maize breeding and precision field experimentation.

Keywords: High-Throughput Phenotyping, maize, plant count, plant height

Acknowledgement: This research was supported by the Science Fund of the Republic of Serbia, Grant No 6672, High-throughput field phenotyping in temperate maize hybrid breeding: how can phenomics improve speed and accuracy of selection? – PHENO_Maize

Application of UAV multispectral and thermal sensing for precision monitoring of maize

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ABSTRACT

The main aim of this study was to investigate the effect of different nitrogen fertilizer levels on maize yield, quality parameters, and canopy thermal dynamics, to determine the optimal phenological stage for yield prediction and get response from the plants during the growing season using UAV based multispectral (NDVI) and thermal imaging. Field experiments were carried out at Látókép experimental station of University of Debrecen under controlled nutrient treatments using multispectral (DJI Mavic 3 M) and thermal sensors (DJI Matrice 4T) with UAV. The relationship between NDVI and grain yield exhibited a clear temporal pattern, with the strongest correlations observed during the growing season. The correlation coefficients reached their maximum (R value: 0.70 - 0.73) between 46 and 73 days after sowing (DAS), particularly under moderate nitrogen supply. Early-stage correlations were low and inconsistent, especially under low nitrogen levels, confirming that early NDVI measurements have limited predictive value usually. Thermal imaging revealed distinct daily canopy temperature dynamics across nutrient levels. Canopy temperatures measured in the early afternoon (2:00PM) showed the highest variability (approximately 34-37.5 °C), with lower nitrogen treatments generally associated with higher canopy temperatures, indicating increased water stress and reduced transpiration efficiency. In contrast, early morning (08:00AM) and evening (6:00PM) measurements exhibited lower temperature ranges (approximately 24-26 °C and 28-32 °C, respectively) and reduced variability among treatments. NDVI measurements are most effective for yield prediction during the intensive vegetative to early reproductive stages (approximately 46-73 DAS), while thermal imaging provides complementary information on plant water status and stress, particularly during midday periods.

Keywords: UAV, maize, long-term experiment, field experiment, multispectral

Optimizing biomethane production from agricultural residues with nanoparticles

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ABSTRACT

Due to the numerous mechanical, thermal and optical properties of nanocellulose and possibilities for future applications, the research of nanocellulose extraction, especially from lignocellulosic biomass, has become very attractive in recent years. Nanocrystalline cellulose has a wide range of applications such as additives in nanocomposite films, use as a template or mold for the synthesis of semipermeable materials and immobilization of proteins. It can also be used to produce smart materials, i.e. materials for the immobilization of bioactive molecules, nanocomposites sensitive to temperature or pH. In addition, previous research has shown that the addition of nanoparticles to an anaerobic digester can increase the efficiency of biogas production. The aim of this research was to isolate the nanocellulosic component (CNC) from the selected biomass (corn, wheat and barley) and determine whether its addition affects the biogas production efficiency. Anaerobic digestion (AD) was carried out without and with the addition of nanoparticles, during which the yield of produced biogas and biomethane was monitored. The highest biogas yield after the addition of nanoparticles was obtained from the corn stover sample, where the maximum value was reached on day 18 of the process and by the end (day 40) it amounted to 155.23 L kg⁻¹ DM, with a biomethane yield of 54 %. The wheat straw sample had a lower biogas yield, with the max. value reached on the day 18 (119.90 L kg⁻¹ DM), with a biomethane yield of 48 %. Based on the research conducted, it can be concluded that the production of biogas and biomethane recorded a slight increase after the addition of nanoparticles. However, to better understand the effect of the addition of nanoparticles on the AD efficiency, it is necessary to conduct further research that will include the addition of different concentrations of nanoparticles in the system.

Keywords: extraction, nanoparticles, anaerobic digestion, agricultural residues

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Biogas Production and Quality from Agricultural Residues

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ABSTRACT

The objective of this study was to evaluate the biogas production potential and quality from different agricultural postharvest residues, including corn, wheat, and barley. Anaerobic digestion experiments were carried out under controlled laboratory conditions to assess methane yield, biogas composition, and process stability. The results showed that maize-based substrates achieved the highest specific biogas yield, reaching up to $0.62 \text{ m}^3 \text{ kg}^{-1}$ volatile solids, with methane content exceeding 55%. In comparison, manure-based substrates produced lower yields but contributed to improved process stability due to their buffering capacity. Co-digestion of crop residues with manure increased overall biogas production by 15–25% compared to mono-digestion systems, while maintaining stable process conditions, with pH values ranging from 6.8 to 7.4 and temperature at $37 \text{ }^\circ\text{C}$. These findings confirm that substrate composition plays a key role in determining both the quantity and quality of biogas. Furthermore, co-digestion represents an effective strategy for improving energy efficiency and ensuring stable anaerobic digestion processes. Future research should focus on optimizing substrate ratios, scaling up the process to pilot and industrial levels, and exploring the integration of biogas production with advanced technologies such as bio-refinery systems. Additional studies are also needed to evaluate long-term process stability and economic feasibility under real operating conditions.

Keywords: biogas, anaerobic digestion, agricultural biomass, methane yield, co-digestion

Acknowledgment: The research was supported by the Croatian-Chinese Scientific and Technological Cooperation 2025-2026, within the project „*Investigation on co-production technology of nanocellulose and bio-methane from lignocellulosic agricultural residues*“

Prediction of Maize Flowering Time Using UAS-Based High-Throughput Phenotyping

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ABSTRACT

UAS-based phenotyping enables efficient acquisition of large-scale field data across genetically diverse plant material, supporting advances in plant breeding. The PHENO_MaizE project focuses on validating digital traits, developing image-based predictive models, and identifying informative variables and optimal flight timings during the growing season. The study includes 400 maize genotypes: 200 inbred lines and 200 hybrids derived as their test-crosses with a single tester. Field trials were conducted at two locations in Serbia in 2026, with UAV-based monitoring performed weekly using a DJI Mavic 3M UAV, generating ~20 time points per trial and location. RGB orthomosaics were produced for all flights and used for analysis. A custom Python-based pipeline was developed to extract image-derived variables and evaluate prediction models for flowering time (anthesis). More than 15 vegetation indices were extracted from each time point and used as model inputs. Several modeling strategies were evaluated to assess the effect of input data selection and trial structure on prediction performance. Models were trained using: (i) variables from all orthomosaics across all time points and both locations; (ii) a subset of five selected orthomosaics per location; and (iii) location-specific datasets, where models were trained and evaluated using data from individual locations only. The same experimental framework was applied to both hybrid and inbred line trials, enabling a direct comparison of model performance across genetic backgrounds. When all orthomosaics were included, Random Forest and Ridge models achieved the highest performance for test crosses ($R^2 = 0.79$ and 0.70). For inbred lines, Random Forest and XGBoost showed the highest accuracy ($R^2 = 0.65$). Future work will assess the robustness and transferability of the models across independent environments and diverse genetic backgrounds.

Keywords: flowering time, high-throughput phenotyping, maize, trait prediction

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Impact of various tires exploitation conditions on the geometric dimensions of their footprints in soil

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ABSTRACT

This abstract contains results of research related to measurements of dimensions of tire footprints in soil. In the experiment two tires: 500/50R17 (radial tire) and 500/50-17 (bias-ply tire) were tested and three levels of inflation pressure (0.08, 0.16, 0.24 MPa) and constant vertical load of wheel (23.5 kN) were used. Research was conducted using the bench which allowed to generate the vertical pressure of the tire on the soil. The research was conducted in static conditions, so the tire footprint was generated in the soil without applying torque. There were no driving or braking forces, which could affect the pressure distribution in the soil. Using the test bench, a tire was loaded with each inflation pressure and a tire footprint was generated in the soil. In next step, a 3D scanner (Smarttech3D Universe) was used to scan the tire footprint. As a result of the scan, a point cloud was obtained that reproduced the shape and geometry of the tire footprint in the soil. Then the mesh of triangles built from the points was created. Then using Smarttech3Dmeasure software the parameters of length, width, depth, and tire-soil contact area (in 3D form) were evaluated. It should be noted that the described parameters were always measured relative to the same measuring point located at the center of the footprint. The real surface was the surface of the footprint in three-dimensional space: not as a simplification in a two-dimensional projection, but as the whole footprint. It was found that the footprint width of both tires increased with decreasing inflation pressure, but values of this parameter were higher for radial tire. In case of footprint depth, it was observed that increase in the inflation pressure resulted in an increase in this parameter for both tires, but significantly higher tire footprint depths were noticed for the bias-ply tire. It should also be emphasized that the tire-soil contact area is the overriding parameter responsible for the distribution of stresses in the soil. It is therefore necessary to continue research in this direction, as this will facilitate the selection of appropriate operating parameters of agricultural tires for soil conditions.

Keywords: tire-soil contact area, soil deformation, 3D scanning, tire footprint

Apple Cultivar Performance Before Agrivoltaic Installation: Baseline Data for Future Impact Assessment

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ABSTRACT

Apples are a globally significant fruit crop, and integrating their production with renewable energy through agrivoltaic (AV) systems presents a novel strategy for sustainable land use. This study, conducted in the inaugural year (2025) of the Agriculture Next Generation (ANG) project in Tenja, Osijek, Croatia, aimed to evaluate the initial pomological characteristics of a diverse range of apple cultivars grown under different AV conditions. The experiment utilized a randomized block design with four replications, comparing three AV treatments (solid, semi-transparent, and dynamic solar panels) against a full-sun control group (Golden Delicious). A total of nine (9) apple cultivars, encompassing summer, autumn, and winter varieties, were assessed. Key pomological traits, including fruit weight, height, circumference, firmness, peduncle length and thickness, and yield, were measured.

The analysis revealed significant differences: Granny Smith and Wilton's Star Red Jonaprince Select produced the largest fruits and largest circumference, while Pinova and Red Topaz had the smallest. Braeburn Lochbuie and Pinova showed the smallest circumference. Red Topaz demonstrated the highest yield, followed by Pinova, while Red Idared showed the lowest productivity. Firmness was greatest in Braeburn Lochbuie and Red Idared, whereas Wilton's Star Red Jonaprince Select had the softest fruits. Peduncle morphology varied considerably, with Sirius showing the highest peduncle volume, while Golden Delicious had notably long peduncles but thin structures.

These initial results from the first year of the ANG project highlight fundamental trade-offs between pomological characteristics and yield potential of nine apple cultivars, establishing a critical baseline for understanding how they will respond to modified microclimatic conditions imposed by overlying agrivoltaic systems in future study years.

Keywords: agrivoltaics, dual-use farming, fruit ecomorphology, *Malus domestica*, renewable energy

Session 6

Youth and Education in Life Sciences

Knowledge and Use of Medicinal Plants among Students of the Faculty of Agrobiotechnical Sciences Osijek

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ABSTRACT

This study aimed to examine the use, perception, knowledge, and educational needs related to herbal products among young adults, with a focus on students. Approximately 60 respondents participated in an online survey, with a high response rate (98.3%). The sample was predominantly female, aged 21–25, mostly undergraduate students, and slightly more represented by individuals from rural areas. The results indicate a high prevalence of herbal product use, particularly in traditional forms such as herbal teas (93.2%) and syrups or honey-based products (66.1%). Essential oils (40.7%) and dietary supplements (39%) were also commonly used. In cases of mild health conditions, most respondents (57.6%) reported combining herbal preparations with conventional over-the-counter medicines, suggesting a complementary approach to treatment. The primary motivation for herbal product use was trust in natural ingredients (42.4%), followed by accessibility (25.4%) and tradition (22%). Herbal products were generally perceived as comparable in cost to conventional medicines. However, safety-related practices were inconsistent, as only 28.8% of respondents always checked dosage instructions and contraindications. Knowledge of medicinal plants was moderate, with most respondents able to identify up to 10 species. Information was mainly obtained from informal sources, particularly family tradition and the internet, while professional sources were less frequently used. Despite this, respondents expressed strong interest in practical education, especially workshops (83.1%) and field-based learning (69.5%). Furthermore, a majority supported increased emphasis on medicinal plants within university curricula. Overall, herbal products are widely accepted and integrated into self-care practices among young adults. The findings highlight the need for improved education and stronger involvement of professional guidance to ensure safe and informed use.

Keywords: medicinal plants, phytotherapy, self-medication, students, health behaviour

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