

Crop Science and Technology: Shaping the Future of Agriculture

International Scientific Conference



BOOK OF ABSTRACTS

September 29 - October 2, 2025 - Belgrade, Serbia

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CROP SCIENCE AND TECHNOLOGY: SHAPING THE FUTURE OF AGRICULTURE

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Snežana Mladenović Drinić, Jelena Vukadinović, Beka Sarić

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Historical overview of maize breeding at Maize Research Institute Zemun Polje

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Maize breeding at Maize Institute "Zemun Polje" began in the 1950s. The collection of initial material and the employment of experts from post-war Yugoslavia was the first step in the implementation of the maize breeding program. The program of creating inbred lines from local populations, started in the early 1950s, marked the beginning of the production of ZP hybrids of the first breeding cycle. Since then, maize breeding has been carried out through seven cycles. Each cycle has been characterized by the introduction of new, potentially more productive hybrids, with improved other agronomic traits (increased tolerance to lodging, lower ear placement, higher seed production, better tolerance to stress factors, etc.). As a result of permanent breeding activities, more than 800 maize hybrids has been registered in Serbia and almost 200 abroad. These results were achieved by the work of generations of breeders and thanks to the training of researchers in the most eminent scientific institutions around the world. The modern breeding program is based on conventional breeding methods, as well as on the introduction of new techniques and methodologies. Specialized machinery have routinely been used for sowing and harvesting for many seasons. Acceleration of breeding process is achieved by use of winter nursery services in South America and by the application of the double haploid technology. Implementation of doubled haploid technology began in 2014, with the aim to create DH lines and new haploid inducers, adapted to temperate climatic conditions. So far, around 20.000 DH lines was created and several maize hybrids, containing at least one DH line have been registered. SNP molecular markers are used for genetic characterization of elite maize germplasm and for the determination of genetic distance between inbred lines. This allows more precise insight into elite germplasm and increases the possibility of creating superior hybrid combinations. Recent research studies are related to the use of high-throughput field phenotyping (HTFP), using RGB and drone-based multispectral cameras for extraction and visualization data per plot. The main idea is to expand knowledge and provide new skills to be utilized for automation of data collection and trait prediction.

Keywords: *maize breeding; breeding cycles; DH lines; SNP markers*

Maize genetic resources: Value for research, breeding and society

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Since the emergence of modern breeding in the mid-1800s, traditional cultivars (landraces) of most regions of the world have been replaced by improved varieties adapted to the objectives of new farming systems. This replacement has been wisely accompanied by the conservation of landraces in genetic resource collections (genebanks) and, to some extent, complementary initiatives such as in situ conservation and participatory breeding. Millions of accessions are currently stored in numerous genebanks throughout the world. They long have been utilised to a very limited extent, to the point that genebanks were called “seed morgues” in the 1980s. I will review how i) the emergence of association genetics gave them considerable value for genetic studies since the early 2000s and how ii) advanced genomic prediction methods now make it possible to incorporate them efficiently in breeding programs to sustain their diversity and long-term efficiency. I will then discuss iii) climate change and agroecological transition call for more diversified varieties, for which genetic resources are expected to be key.

Keywords: *landraces; climate change; genomic prediction; crop diversity*

Is phenomics a good investment for crop improvement? A Texas maize breeders' perspective

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High-throughput remote sensing platforms such as near infrared reflectance spectroscopy (NIRS) and drones (unoccupied aerial systems, UAS or UAVs) are inexpensive, scalable, and reliable to collect phenotypic measurements on crops. Such measures go beyond conventional phenotypes to include high-dimensional previously unmeasurable phenomic features, including temporally over crop growth. Statistical methodology (mixed models, machine learning, deep learning) demonstrates novel phenomic features extracted from remote sensing tools are heritable and predictive. Our research and others have shown that phenomic approaches complement or can even replace genomic approaches in objective prediction of agronomic end phenotypes such as grain yield, disease, lodging, senescence and nitrogen. Phenomic features uniquely incorporate most genetic (G, including additive, dominant, epistatic), environment (E), and GxE interaction variance. Therefore, phenomic predictions tend to be more useful across diverse populations and environments than genomic markers. A disadvantage of phenomic approaches is that methodology and software are where genomics was 25 years ago. Therefore, specialized expertise is still needed to operationalize these tools within a breeding program, along with management of the massive data. Additionally, more phenomic features are still needed along with methods to integrate sparse data across environments. Our group has developed multiple methods, packages and scripts to help lower these barriers. Numerous ancillary benefits have also been found from phenomic data collection, including novel biological findings, novel trait discovery, prediction of data from lost plots or locations, and ability to visualize plots from past years throughout their growth. Given the low costs and ease of data collection, especially with drones, all breeding programs would benefit from routinely collecting temporal image data immediately; these specific environments will never be available again, and the ancillary uses can benefit breeding and research programs today. However, it is expected that drone-based phenomic selection will not become routine for a few years.

Keywords: *drone; phenotyping; phenomics; breeding; maize*

Acknowledgment: USDA-NIFA; FFAR; USDA-ARS; Texas Corn Producers Board; Eugene Butler Endowed Chair.

Rapid population improvement using fertile and vigorous haploids in maize

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Homozygous and homogeneous doubled haploid (DH) lines can be obtained in two generations, which - in maize - requires haploid induction and subsequent doubling of genomes to transition from haploid to diploid plants. Sterility and reduced vigor are two major limitations in working with haploid plants. We refer to the weaker performance of haploids vs. isogenic diploids as “haploid frailty”. The extent of haploid frailty can be quantified for any trait as a percentage reduction in performance of haploids versus isogenic diploids. In a GWAS panel of DH and derived haploid isogenic lines, haploids displayed reduced performance compared to diploids, particularly for plant and ear height (30-40% reduction). Heritabilities were high for diploids (0.8 to 0.9) and slightly lower for haploids (0.7 to 0.9), while haploid frailty heritabilities ranged from 0.1 to 0.7. Positive correlations between diploid and haploid traits suggest that diploid performance may effectively predict haploid performance, and vice versa. Conversely, haploid performance correlated negatively with haploid frailty, particularly for ear and plant height, suggesting that selecting vigorous haploids reduces frailty and enhances haploid female fertility (HFF). It is conceivable that vigorous and fertile haploid plants can be generated, enabling rapid two-generation breeding cycles that alternate between crossing fertile haploids and inducing resulting heterozygous diploids to return to fertile haploids.

Keywords: *doubled haploids; haploid male fertility; haploid female fertility; haploid frailty; population improvement*

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Agronomic and genetic approaches, including CRISPR, for producing low asparagine wheat, in the context of evolving regulations on genome edited crops and acrylamide in food

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Acrylamide is a carcinogenic processing contaminant that forms from free (non-protein) asparagine and reducing sugars during high-temperature cooking and processing of cereal grains and other plant products. Food businesses face the prospect of the European Union setting Maximum Levels for acrylamide in food this year. Compliance would be greatly facilitated by the development of crop varieties with reduced acrylamide-forming potential. We have used CRISPR/Cas9 to knock out the asparagine synthetase-1 and -2 (TaASN1 and TaASN2) genes of bread wheat cv. Cadenza. Mutations in the A, B and D genome TaASN2 genes have also been produced by chemical mutagenesis and stacked to produce a total TaASN2 knockout of cv. Claire. We have also assessed the effect of a natural deletion of B genome TaASN2. In field trials, the CRISPR lines show significant (up to >90%) reductions in free asparagine concentration and a concomitant decrease in acrylamide formation in heated flour, biscuits and bread, especially after toasting. These genetic approaches are being undertaken alongside experiments on fertilisation rates, especially of sulphur, and disease control, with the signalling hub through which pathogen infection induces free asparagine accumulation being elucidated in detail. We are also attempting to add a high lysine trait to the low asparagine wheat. The work is being conducted during a period of rapid change in regulations governing genome edited crops in the UK, culminating in the Genetic Technology (Precision Breeding) Act (March 2023), and its associated secondary legislation (March 2025). The UK is expected to start processing applications to market precision bred organisms (PBOs; GE crops with no transgene present) in November 2025. The EU is also moving to change its regulations on GE crops through the development of its New Genomic Techniques legislation. The acrylamide issue in cereal products is the subject of the ACRYRED COST Action (<https://acryred.eu/>).

Keywords: *wheat acrylamide; CRISPR/Cas9 Asparagine/Lysine; food safety; nutritional enhancement*

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Separating genetic and environmental impacts on the maize microbiome

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Microbes live on, around, and inside maize, yet we know little about how they impact the plant. Two key questions are how the local environment and host genetics shape these microbial communities, and what sorts of interactions happen between them. To address these questions, we performed two analyses of maize microbiomes across the United States as part of the Genomes to Fields Initiative. Our first analysis focused on soil and endophyte microbiomes from young (<1 month old) plants at 30 locations. We confirm that the plant acts as a strong filter, with endophyte communities severely restricted relative to soil and rhizosphere ones. We also find that endophyte communities are much more interconnected than soil and rhizosphere communities, with many more microbe-microbe interactions present. We find few consistent effects from specific environmental variables (pH, nutrient levels, etc.), and soil and rhizosphere communities are actually more consistent across locations than endophyte communities. Our second analysis focused on stalk endophytes in 20-30 hybrids at 20 locations, sampled at anthesis. We find that host genetics by itself has almost no consistent effect across environments, yet gene-by-environment interaction is substantial. This pattern holds across multiple measures of microbial diversity and is consistent with previous data from the maize rhizosphere. These results imply a 2-step model where the environment determines the initial diversity that is then modified by host genetics. This modification is not consistent across environments, implying there is no “ideal” microbiome for a genotype. We do find associations between the stalk endophyte community and environmental conditions (especially soil potassium), which merit further investigation. Taken together, these results indicate that the maize microbiome is a complex trait subject to strong gene-by-environment interaction, and this fact needs to be taken into account when designing future experiments.

Keywords: *maize; microbiome gene-by-environment interaction, endophyte; quantitative genetics; heritability*

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High-throughput plant phenotyping in the era of LLMs: What to expect in the coming years

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For more than a decade, Hiphen has been at the forefront of high-throughput plant phenotyping, delivering solutions for field trials, post-harvest assessments, and greenhouse experiments. This period has seen a wave of technological breakthroughs: drones have made large-scale, high-resolution data collection faster and cheaper; deep learning has unlocked the extraction of complex traits once considered out of reach (such as accurate plant counts); and predictive breeding — extending genomic prediction to the realm of phenotyping — now integrates environmental interactions (G×E) to accelerate genetic gain. We are now entering a new disruptive phase driven by large language models (LLMs) like ChatGPT, Claude, and Mistral. Beyond their conversational abilities, these AI systems can streamline phenotyping workflows, enhance data interpretation, and open entirely new research and breeding applications. In this presentation, we will explore today's cutting-edge tools for phenotyping crops such as corn, and look ahead to how LLMs could reshape the way we design experiments, process images, and translate phenotypic data into actionable insights in the years to come.

Keywords: *predictive breeding; AI in agriculture; drones; large language models (LLMs); data interpretation*

A novel tool for longitudinal phenotyping in maize: Towards the „clinical trials“ in agriculture

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The success of maize breeding programmes relies on trustworthy, high resolution information from multilocation field trials. Secondary traits frequently show greater heritability than grain yield, and monitoring their trajectories throughout the season (longitudinal phenotyping) exposes stress episodes weeks before harvest. However, inconsistent instrumentation and protocols make cross site standardization difficult. We approached this challenge by fusing two complementary data streams: sub centimetre scale UAV imagery captured with low cost multispectral cameras and freely available Sentinel 2 satellite products (~10-30 m pixels) accessed through Google Earth Engine and Copernicus Hub. A key innovation is an adaptive over and undersampling routine that spatially resamples both datasets toward a common target resolution. This bidirectional resampling, combined with radiometric normalisation, produces a fully cross calibrated longitudinal reflectance series in which every pixel is temporally and spectrally comparable, an advantage unavailable in current breeding pipelines. On these harmonised stacks we built an open source workflow that operates in sliding temporal windows. For each plot, the pipeline interpolates reflectance, integrates vegetation indices (NDVI, NDRE, GI, red edge, PRI, etc.), masks non vegetation pixels and feeds the resulting features into gradient boosted models for grain yield prediction. In trials with 128 hybrids grown under rain fed temperate conditions, window integrated indices explained up to 63 % of yield variance, more than doubling the explanatory power of single date snapshots at flowering or maturity. The calibrated, dense data capture also offered earlier and more reliable warnings of performance divergence, enabling in season management decisions. All core modules such as sensor calibration, bidirectional resampling, masking, temporal interpolation, plot demarcation and model training will be released under an open-source licence during the PHENO_MaizE project. By delivering economical, high throughput and fully calibrated longitudinal phenotyping, the tool brings field trials closer to the rigor of medical “clinical trials” and accelerates maize variety development across public and private sectors.

Keywords: UAV; phenotyping; multispectral; temporal; spatial

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Integrating high-throughput field phenotyping into maize breeding PHENO_MaizE project achievements and preliminary Results

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Climate change, population growth, and global instability continue to challenge food production systems, calling for faster and more accurate plant breeding strategies. Accurate and efficient phenotyping remains one of the major bottlenecks in modern plant breeding, especially for crops like maize that are widely cultivated and genetically diverse. The PHENO_MaizE project explores the use of high-throughput field phenotyping (HTFP) using RGB drone imaging to support selection decisions in hybrid maize breeding programs under temperate conditions. Across three environments in Serbia, RGB images of 200 inbred lines and their test-crosses to a common elite tester are captured up to 30 times during the growing season. Each image set is used to generate orthomosaics and digital surface models (DSMs), from which approximately 30 phenotypic features—including plant height, canopy cover, plant count, and vegetation indices—are extracted. The study has three major goals: (1) evaluating the potential of image-derived traits (such as digital plant height and plant count) to replace manual measurements; (2) developing prediction models for flowering time, grain yield, and grain moisture at harvest using image features and machine learning methods; and (3) identifying the most informative image-derived variables and flight time points for prediction. Additionally, we explore an innovative pipeline where entire orthomosaic images are used directly as input for deep learning models, bypassing the need for trait extraction. Our approach targets practical application in small to medium-sized breeding programs with limited resources. Through this work, we aim to increase the efficiency, accuracy, and scalability of phenotyping in maize breeding. By identifying critical time points and informative variables for selection, the project provides a framework for integrating HTFP into routine breeding workflows, ultimately enhancing genetic gain.

Keywords: *high-throughput phenotyping; UAV; maize breeding; RGB imaging; machine learning*

Acknowledgment: 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.

Molecular strategies for thermotolerant pollen: A translational perspective

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Pollen germination and pollen tube growth are among the most heat-sensitive stages of plant reproduction. High temperatures induce a global downregulation of translation while promoting the selective synthesis of proteins required for cellular survival and protection against protein misfolding. In *Arabidopsis thaliana*, the eif3m1/eif3m2 double mutant exhibits impaired pollen germination, compromised tube integrity, and increased seed abortion even under normal conditions. Unexpectedly, however, eif3m1 single mutants display improved pollen germination and enhanced tube stability under heat stress, outperforming the wild-type Col-0. This thermotolerant phenotype is associated with the upregulated expression of the eIF3M1 paralog eIF3M2, which drives elevated HSP70 mRNA and protein levels. Overexpression of eIF3M2 enhances HSP70 expression, while its knockdown reduces HSP70.1 promoter activity and increases pollen tube bursting under elevated temperatures. Moreover, eIF3M2 physically interacts with both HSP70 and eIF4G proteins. Together, these findings identify an eIF3M2–HSP70 regulatory module as a key determinant of pollen tube thermotolerance, preserving membrane integrity and supporting fertilization and seed set under heat stress. This mechanism reveals a novel strategy by which plants safeguard reproductive success under climate-induced temperature extremes.

Keywords: heat stress; thermotolerance; climate change; protein translation; fertilization

SNP analysis for maize breeding progress in Ukraine

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Global maize breeding widely employs molecular markers, and SNP-based selection is rapidly developing in Ukraine. SNP genotyping has been conducted on 2,161 Ukrainian and global maize inbred lines using the Illumina GoldenGate platform (BDI III panel, 384 markers). Processing the SNP data has provided a theoretical basis for their use in routine breeding. This includes the estimation of SNP allele frequencies and pairwise genetic distances, analysis of the genetic structure of Ukrainian maize lines based on ancestral population contributions, regression analysis of SNP-associated traits within diallel schemes, and STRUCTURE clustering. As a result, an effective SNP implementation technology was developed. SNP analysis enhances modern breeding efficiency by enabling accurate identification of genetic resources, monitoring inbreeding, and predicting heterosis potential. Since 2011, molecular markers have been integrated into Ukrainian breeding programs. Hybrids and inbred lines registered after 2015 have been developed using SNP technologies. Between 2015 and 2025, 56 heterotic hybrids and 61 inbred lines were patented and included in the Ukrainian Variety Register. In the EU Common Catalogue, 28 hybrids are listed: 26 in Romania and one each in Slovakia and Lithuania. Furthermore, functional markers such as crtRB1-3'TE revealed allelic variation associated with provitamin A content in grain, demonstrating potential for marker-assisted biofortification. These findings support the utility of combining neutral and functional SNP markers to improve agronomic traits and nutritional value, particularly within precision breeding frameworks. The next step in the development of SNP-associated technologies should be the creation of new-generation panels that integrate functional markers of economically valuable traits with markers for variety certification and genetic clustering. This study highlights the importance of large-scale genotypic databases and their use in hybrid prediction and diversity conservation. The implementation of SNP technologies in Ukrainian maize breeding offers a scalable, data-driven approach to strengthening food security, export competitiveness, and climate resilience.

Keywords: *Zea mays L.*; molecular markers; SNP analysis; heterosis; breeding

The effect of low temperatures on the maize transcriptome during the emergence stage

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Maize is a crop plant of great importance for agriculture; however, it is extremely sensitive to negative environmental factors caused by climate change. Consequently, developing strategies to mitigate the effects of climate change on the production of maize is a matter of urgency. Early sowing in temperate areas is a strategy that ensures avoiding the exposure of maize plants to high temperatures and drought during the most sensitive growth stages (flowering and grain filling). However, early sowing also results in exposure of maize seedlings to low temperatures (LT) during the emergence stage. For that reason, it is necessary to fully comprehend the mechanisms and pathways responsible for establishing LT tolerance in the early developmental stages of maize. For this purpose, five-day-old maize seedlings belonging to two maize lines of contrasting tolerance to LT (LS and LT) were subjected to low-temperature treatment (10/8°C), after which whole transcriptome sequencing was performed. Sampling was done after 6h and 24h of LT treatment, and the same was done for control seedlings grown in optimal conditions (25°/20°C). The sequence data was then used for expression analyses specific for different RNA classes – messenger (genes), lncRNA, circRNA, and miRNA. The results showed that 508 genes, 63 lncRNAs, and 649 miRNAs were potentially involved in the LT-response in maize during the emergence stage, while circRNAs did not seem to be included in the response. Low temperatures appeared to negatively affect photosynthesis and protein stabilization. Additionally, the two genotypes showed differing expression patterns of genes and ncRNAs involved in these processes, suggesting that these RNAs could be important for the establishment of LT tolerance.

Keywords: maize; emergence stage; whole transcriptome profiling; low temperature stress

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Mitigation of mycotoxin contamination along the cereal food chains: Advanced strategies in the current climate crisis

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Mycotoxins, secondary metabolites produced by toxigenic fungi, are natural contaminant of food and feed and represent a serious threat worldwide, because of their impact on animal, human and plant health. According to a recent report carried out by Eskola et al. (2020), around 25% of food is contaminated by at least one mycotoxin, with important implications for humans and animals health and global trade and economy. Cereals are among the most important commodities worldwide for consumption, economic relevance, as well as susceptibility to mycotoxin contamination. Main fungal genera which species cause mycotoxin accumulation in the field are *Aspergillus*, and *Fusarium*, although also *Alternaria* can include harmful species. Hot and dry weather conditions, prolonged periods of rainfall deficits, and tighter supplies and market uncertainty have contributed in the last years to reduce cereal production and increase the risk of mycotoxin contamination worldwide. Therefore, mycotoxin management in these crops requests advanced strategies along the whole supply chain, from pre- to post-harvest stages. Adopting an integrated pre- and post-harvest approach, possibly using different reduction techniques, still remains the key action to manage mycotoxin contamination risk at a global level. In this presentation, an overview of advantages and disadvantages of the different approaches to reduce mycotoxin contamination in cereals through pre-and post-harvest practices will be given, ranked for importance and urgency of action.

Keywords: food safety; global trade; post-harvest management; *Aspergillus*; *Fusarium*

Herbicide Tolerant Maize and Soybeans in USA: 25 years later

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Overwhelming percent (>90%) of maize and soybean acres in USA are planted with herbicide tolerant crops (HTC), including: Roundup-Ready (glyphosate), Liberty-Link (glufosinate), Xtend (dicamba) and Enlist (2,4-D). Objective of this overview was to provide a history, and major advantages and disadvantages of widespread use of HTCs in the last 25 years. Advantages include: (1) broadened spectrum of weeds controlled, (2) increased crop safety, (3) reduced risk of herbicide carryover, (4) crop management simplicity. Major disadvantages include: (1) risk of single selection pressure, especially with repeated use of glyphosate, (2) shifts in weed species, (3) gene flow and contamination of organic crops, (4) herbicide drift. It is easy to fall into a trap of overusing single herbicide (glyphosate), when one glyphosate-tolerant crop was grown after another, due to extensive marketing by the companies that sell this technology. This misuse of glyphosate resulted in resistance in 18 weed species. Especially problematic ones are the two *Amaranthus* species, which rendered RR technology almost useless in fields infested with glyphosate-resistant Palmer amaranth (*A. palmeri*) and common waterhemp (*A. rudis*). Therefore, I believe that the further use of these crops should be only within the concept of integrated weed management, which is the key to preserving the longer-term benefits of this technology, while avoiding many of the concerns about their use, or misuse.

Keywords: *biotechnology; herbicide tolerant crops; weeds*

Maize (*Zea mays* L.) hybrids response to relative time of the emergence of weeds and their weed competitive ability in a Mediterranean environment

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The timing of weed emergence in relation to the crop can determine the impact of weed interference on crop yield performance especially in the semi-arid environments of the Mediterranean region. Field trials were conducted in western Greece to evaluate the effects of the timing of weed emergence on the yield of maize hybrids (*Zea mays* L.) as well as on weed growth and fecundity. In the first trial, barnyardgrass (*Echinochloa crus-galli* (L.) P. Beauv.) was the weed species under study that emerged at the VE, V2, V4 and V6 growth stages of maize. Barnyardgrass plants that emerged at the VE stage produced 1010–1305 seeds plant⁻¹ compared to only 112–240 seeds plant⁻¹ produced from plants that emerged after the V4 stage. Maximum grain yield losses ranged from 24 to 34% for early-emerging barnyardgrass while less than 9% yield loss occurred for later-emerging seedlings. The hybrids Factor and Dracma showed the fastest early growth rates and highest competitiveness. In the second trial, the interference between velvetleaf (*Abutilon theophrasti* Medik.) and maize was investigated. Velvetleaf emerged at the growth stages V1, V3, V5 and V7 of maize. Velvetleaf that emerged at the V1 growth stage produced up to 17 times less seeds than plants that emerged at the V5 growth stage. The maximum yield loss was between 26 and 37% for early emerging velvetleaf, and less than 6% for seedlings emerging at the V7 growth stage. At 40 days after sowing, the relative growth rates for the maize hybrids Pako, Mitic, Agrister and Arma were 1.31, 1.48, 1.05 and 1.75 cm plant⁻¹ day⁻¹, respectively. The results suggest that the selection of competitive maize hybrids and the prevention of early weed interference are essential to avoid severe grain yield losses in maize fields in the dry Mediterranean environments.

Keywords: maize hybrids; weed competitive ability; time of emergence

Advancing precision application science through MRIZP and PAT lab collaboration

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Established in 2012 within the historic West Central Research, Extension and Education Center (WCREEC, founded in 1904), the Precision Application Technology (PAT) Lab serves as a national leader in promoting safe, effective, and environmentally responsible pesticide use. Building on over four decades of pesticide application research, the lab supports the optimization of approximately 900 million pounds of active ingredients applied annually across U.S. cropland. By integrating advanced nozzle technologies, application parameters, and sprayer modifications, the PAT Lab develops science-based solutions to enhance pesticide efficacy and minimize drift and off-target movement potential. The PAT Lab's research spans laboratory, greenhouse, and field environments, investigating how nozzle selection, spray quality, solution characteristics, and pressure influence efficacy and crop safety. Its mission also encompasses translating these findings through outreach and education, empowering producers with knowledge to implement best management practices that improve agricultural sustainability. In alignment with this mission, the collaboration between PAT Lab and the Maize Research Institute "Zemun Polje" (MRI) has flourished since 2019 under the joint leadership with Dr. Milan Brankov. This partnership has yielded impactful publications on adjuvants, drift reduction, herbicide programs, and agroecological practices, highlighting a mutual commitment to advancing practical, science-backed solutions. In 2025, the collaboration expanded to include research on newly registered conventional and OMRI-listed products, evaluating their efficacy, crop safety, and environmental behavior. Beyond joint trials, additional collaborations emerged following visits, including contributions to data analysis, manuscript and project(s) development. These efforts aim to ensure practical relevance, particularly in an era when evolving regulatory frameworks require thoughtful and impactful research. Together, MRI and PAT Lab are shaping the future of sustainable crop protection.

Keywords: *drift reduction; nozzle technology; adjuvants; sustainable agriculture; pesticide application*

Impact of nanostructured nitrogen fertilizer "nano urea" on morpho-agronomical traits of common wheat (*Triticum aestivum* L.)

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Common wheat (*Triticum aestivum* L.) is one of the leading crops in the Republic of Slovenia. In the sowing structure of the Republic of Slovenia, it occupies approximately 30,000 hectares every year. It is known that small cereals such as common wheat are a large consumer of mineral nitrogen fertilizers, such as calcium ammonium nitrate, which contains 27% nitrogen. In the past, nitrogen mineral fertilizers were most often discussed through their negative impact on the environment. After 2022, however, they are discussed mainly from the perspective of how to reduce the dependence of Slovenian agriculture on the import of mineral nitrogen fertilizers with alternative methods in plant nutrition. In the research work, we focused on the issues related to the use of mineral nitrogen fertilizers in the production of common wheat. The sciences about nano fertilizers represents an important field that can offer solutions to problems related to the production and use of conventional mineral nitrogen fertilizers. Nano-fertilizers offer numerous advantages over "conventional" fertilizers, such as greater efficiency in nutrient absorption by plants, absence of volatilization, and prolonged action. The research work carried out can confirm the feasibility of using nano-structured nitrogen fertilizers such as "nano urea" in the production of common wheat.

Keywords: *common wheat; morpho-agronomical traits; nano urea; nitrogen; variety*

What we learned so far considering acrylamide in thermally processed foods?

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In April 2002, researchers in Sweden showed that acrylamide, a potential carcinogen for humans, forms during heating of certain foods at high temperatures and low moisture conditions associated with frying, baking, and roasting. The discovery of acrylamide in carbohydrate rich cooked foods raised worldwide concern among consumers and initiated intense scientific research into possible measures to control the formation of this compound in thermally processed foods. As a probable human carcinogen, acrylamide is formed when food is heated to high temperatures exceeding 100°C and is thus considered a naturally occurring compound. After its discovery, acrylamide has been one of the hottest topics of food science. The Maillard reaction between asparagine and carbonyl compounds, such as reducing sugars, forms acrylamide in foods during heating. Foods high in asparagine and reducing sugars, like those containing aldose and ketose sugars, are most susceptible to acrylamide formation. Low free asparagine cereal varieties and flours, like rice or maize, are key to reducing acrylamide in bakery products. Avoiding reducing sugars and ammonium bicarbonate in recipes can also help limit acrylamide formation. Acrylamide, found in various cooked foods, is primarily sourced from fried potato products, bakery foods, and coffee. Exposure levels vary based on food type and processing, with fried potato products being a significant contributor for both adults and children. The EU legislation sets benchmark levels for acrylamide in food, requiring food businesses to demonstrate good manufacturing practices and take mitigation actions to reduce acrylamide content. Food businesses should be aware of potential acrylamide regulations and ensure staff are familiar with ALARA principles to minimize levels. Mitigation can be achieved by modifying processing conditions and/or product formulations. The Acrylamide Toolbox, released in 2019, provides mitigation strategies for reducing acrylamide in food products. While there is no single solution, product development and consideration of ingredient choices are crucial for maintaining consumer perception and reducing acrylamide levels.

Keywords: *acrylamide; formation mechanism; mitigation; cooked foods*

Acrylamide in cereal snacks and possibilities for its mitigation

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Acrylamide is a process contaminant that is formed during the heat treatment of plant-based foods containing saccharides and the amino acid L-asparagine, and therefore cannot be avoided in heat-treated cereal foods. Foods made from cereals are an important source of energy and biologically valuable substances, and products such as bread and baked goods are staple foods with significant consumption. In an effort to increase the attractiveness of cereal products, nutritionally valuable sources are used in the design of new cereal products and new forms of cereal products are being developed, especially long-life products with a low number of ingredients and a simple recipe. Such foods include puffed single- or double-ingredient snacks using nutritionally valuable cereals and legumes, which, however, have an increased potential for acrylamide formation. The results of monitoring the acrylamide content in long-life puffed snacks, as well as the possibilities for its elimination and practical experience in applying acrylamide mitigation procedures, are the subject of this lecture.

Keywords: *acrylamide; cereal-based products; legumes; asparagine; snacks*

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Exploring the bioactive compounds of ZP cereals and legumes and their potential applications: Bridging food science with modern breeding

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Regardless of the nation or dietary preferences, cereals and legumes have been among the indispensable staple food ingredients for centuries. These grain crops contain a considerable quantity of bioactive compounds that act as antioxidants and can potentially benefit human health in numerous ways. The latest advancements in crop breeding ensure that these grain crops can adjust to changing environmental conditions while maintaining their nutritional value and a wide range of health-promoting components. The objective of this study was to investigate the contents of bioactive compounds of ZP cereals and soybean genotypes in order to assess the potential of their application. By including the byproducts of grain processing and agricultural residues such as fine powder residue from blue popcorn milling, corn silk, oat husks, and pigmented soy hulls into research, this study also aimed to decrease agricultural waste and help increase the sustainability of the agri-food sector. The main bioactive compounds detected in the studied cereals and legumes were phenolic compounds (phenolic acids, flavonoids, anthocyanins), carotenoids, tocopherols, and dietary fibers (β -glucans). The observed applications included functional shortbread cookies and pickled baby corn enriched with anthocyanins from black soybeans, confectionary spreads from sweet corn residues and black soy hulls, microencapsulates of blue maize polyphenolics, nutraceutical herbal extracts (corn silk tea), and anti-aging skincare products (skin cream with antioxidants). The observed applications demonstrate the versatility and potential of the grain-derived bioactive compounds in various industries. Overall, the intersection of research and practical application in this field holds significant promise for creating a more sustainable future while maximizing the value derived from agricultural processes.

Keywords: *cereals; legumes; bioactive compounds; potential application; sustainable crops*

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FAST tracking; Simulating reproductive thermotolerant under the microscope

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Advances in molecular research are accelerating the discovery of thermotolerant traits in plants, outpacing traditional reliance on natural hybrids. Natural hybridization provides extremely valuable genetic diversity but is inherently slow with the current rate of climatic changes, constrained by reproductive incompatibility and random genetic recombination. In contrast, molecular approaches such as genomics, transcriptomics, and CRISPR-based gene editing enable precise identification and manipulation of genes and regulatory pathways linked to heat tolerance. These tools not only uncover novel allelic variants and stress-responsive networks but also allow for the rapid validation and transfer of thermotolerance traits across species barriers among crops. By integrating functional assays, we can construct detailed molecular maps of stress adaptation, guiding targeted breeding and synthetic biology strategies. In this meeting, I will highlight our new approach in simulating environmental heat stress at the cellular level in combination with high resolution imaging to study reproductive thermotolerant with the goal of accelerating discovery of useful reproductive heat resistant traits. By combining traditional diversity with molecular precision, these approaches offer faster, more scalable solutions for developing crops adapted to the challenges of climate change.

Keywords: *molecular breeding; transcriptomics; high-resolution; imaging CRISPR gene editing; climate change*

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From seed science to ISTA rules for seed testing

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ISTA's vision "Uniformity in seed quality evaluation worldwide ", having as objectives to adopt standard procedures for sampling and testing and to promote research in all areas of seed science and technology. The ISTA test methods are based on scientific knowledge and the accumulated experience of those working in seed testing and quality control. This expertise is provided by the members of ISTA's Technical Committees. All test methods proposed for inclusion in the ISTA Rules have gone through the ISTA Method Validation Programme. To stay updated, ISTA is revising its Rules annually. A positive impact on food and feed supply can be achieved by improved seed breeding processes. They always require high seed quality, which can only be achieved by uniform seed testing. Seed quality is influenced by several factors and ISTA plays an important role in maintaining and improving testing methods. Seed is a living biological organism, and its behavior cannot be predicted with the certainty that characterizes the testing of inert, non-biological material. Characteristics such as trueness to variety and or genetic purity, germination, physical purity, viability, vigour, are important aspects for breeders, seed producers, and farmers as final users of the seeds. The above-mentioned characteristics and not only will lead to a uniform crop and successful establishment and support to avoid weedy, diseased crops and to avoid uneven maturity. Among the most relevant aspects, mechanical damage may affect seed germination, health, and vigour. One of the strategic goals of ISTA is to develop tests and methods based on science and new technologies and facilitate the application of new or emerging technologies into current methods. Some achievements are the introduction of the DNA-based methods for variety testing, X-radiography, vigour tests and GMO testing (detection, quantification, or confirmation of the presence of genetically modified organism seeds).

Keywords: *seed; seed quality; testing methods; new technology; ISTA*

The ECPGR European Evaluation Network EVA, successful public-private partnerships for evaluation of European PGRFA

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The European Cooperative Programme for Plant Genetic Resources (ECPGR) is a network of most European countries aimed at ensuring conservation and sustainable use of plant genetic resources for food and agriculture (PGRFA) in Europe through collaborative activities. Since 2019, the ECPGR initiative European Evaluation Network (EVA, <https://www.ecpgr.org/eva>) brings together diverse stakeholders in public-private partnerships to jointly evaluate vegetable and field crop accessions conserved in European genebanks, which are often poorly characterized and consequently underutilized. At present, more than 130 partners including genebanks, research institutions, private breeding companies and farming cooperatives from 34 countries collaborate in six networks on carrot, lettuce, pepper, grain legumes, maize, wheat and barley, and an EVA network for perennial crops is in preparation. So far, EVA networks have generated valuable standardized evaluation data for priority traits on >5,000 PGRFA accessions, which together with genotyping data allowed analyses of genetic diversity and marker-trait associations. The EVA Maize network was established in 2020 and currently has 24 partners from 13 countries, including 9 private companies. During the first phase of the project the network jointly evaluated >600 European maize landraces and 360 testcross populations in 90 trials across 30 locations in 10 countries, generating multilocation phenotypic data. Genotyping of the EVA Maize collection revealed a large diversity and identified previously unstudied genetic groups which could be useful for enhancing the genetic basis of breeding germplasm. Leveraging their collaboration, the network partners engaged in complementary projects MineLandDiv and MalaNIRs, which provided the opportunity to extend the geographic scope of the network and its collection, enhancing also the benefits for participants. Here we present an overview of the EVA framework, results from and an outlook for the EVA Maize network, as an example for a successful public-private partnership promoting sustainable use, presenting perspectives for improved management and valorization of PGRFA.

Keywords: *characterization; evaluation; genetic resources; public-private partnerships*

Acknowledgment: EVA is funded by BMEL, Germany (GenRes 2019-2 and 2024-2), the AGENT project (H2020 grant 862613) and ECPGR.

CREDIT Vibes project: From science to innovative agrotechnology through *Trichogramma*-based biocontrol in maize

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The CREDIT Vibes project (Horizon Europe, GA 101059942) builds a cross-border ecosystem for sustainable crop protection, bringing together researchers, SMEs, and farmers to accelerate the adoption of green-editing, biological, and digital innovations. A flagship success of this collaborative effort is the development and deployment of *Trichogramma*-based capsules for biological control of the European corn borer (*Ostrinia nubilalis*), one of the most damaging maize pests, responsible for yield losses of up to 20% annually. In 2024, the technology was tested at three locations - Novi Sad, Kikinda, and Zemun - covering a total of 16 hectares. Using DJI Matrice 300 RTK drones equipped with specialized applicators, 2,000 capsules containing *Trichogramma* parasitoids were released, resulting in the deployment of approximately 4 million beneficial wasps. This precision, drone-based application ensured uniform field coverage, significantly reduced pest pressure, and minimized labor costs and environmental impact. Results highlight the potential of *Trichogramma*-based capsules as a scalable, environmentally friendly alternative to chemical pesticides, directly supporting the EU Farm to Fork strategy and the Green Deal objectives. In 2024, pre-treatment monitoring showed that in maize hybrid I, 82% of eggs were non-parasitized and only 18% parasitized, while in hybrid II, 100% of eggs were non-parasitized. After capsule application, parasitism rates increased: in hybrid I, non-parasitized eggs dropped to 4% and parasitized reached 96%, and in hybrid II, parasitism rose from 0% to 86%. These results demonstrate strong pest suppression capacity and validate capsule deployment as a highly effective and replicable biological control tool. By integrating drone technology with biological control, CREDIT Vibes provides a model for sustainable pest management that can be scaled across Europe. In 2025, the technology was scaled to commercial fields by Al Dahra Serbia, demonstrating a remarkable achievement of direct technology transfer from research to practice within one season. This case highlights how CREDIT Vibes transforms EU-funded research into tangible impact, strengthening food security, sustainability, and innovation in maize production systems. Through this example, CREDIT Vibes illustrates how EU collaborative projects can bridge research excellence and field-level practice, advancing the European Green Deal, Farm to Fork Strategy, and Mission on Climate-Neutral Cities.

Keywords: climate-smart agriculture; farm to fork strategy; beneficial insects; drone application; green deal

Practical breeding methods: Example of a common bean breeding program from Slovenia

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Slovenia has a long tradition of cultivating common beans (*Phaseolus vulgaris* L.), evidenced by the many local varieties. These varieties are primarily cultivated on small-scale farms and in gardens using low-input production systems. The Slovenian national common bean breeding program, led by the Agricultural Institute of Slovenia, exemplifies a comprehensive, data-driven approach to improving legumes. The program combines traditional selection methods with modern molecular tools to improve traits such as yield, nutritional quality, disease resistance, and adaptability to challenging agro-climatic conditions. Targeted hand-pollination of snap and dry beans has been successfully applied. This involves crossing elite parental lines and selecting based on observations and evaluations during the growing seasons, such as plant growth and development, resistance to pathogens and pests, and tolerance to abiotic stress, as well as technological maturity, including morphological characteristics, yield, and seed quality. To accelerate the breeding process efficiently, MAS (marker-assisted selection) was implemented for selected agronomic and nutritional quality characteristics using trait-related DNA markers. Results from MAS applied in the F₂ generation of selection and self-pollination are combined with phenotypic selection and other in vitro and biochemical analyses. The overall data are then used to calculate key breeding parameters, which are essential metrics and concepts used in plant breeding to evaluate and improve the genetic traits of interest. This program highlights the integration of traditional selection methods with modern data analytics to enhance genetic diversity, improve agronomic performance, and support sustainable agriculture. This program provide a solid foundation for developing high-yielding, nutritious, and locally adapted common bean varieties in Slovenia and beyond.

Keywords: *crop improvement; agronomically important traits; hand-pollination; trait-related DNA markers; key breeding parameters*

Acknowledgment: This program is supported by the Slovenian Research and Innovation Agency – ARIS (former Slovenian Research Agency – ARRS (Javna agencija za raziskovalno dejavnost Republike Slovenije)) under the Agrobiodiversity Research Program [P4-0072], the bilateral project between Slovenia and Serbia [BI-RS/23–25-042], and the Slovenian Ministry of Agriculture, Forestry and Food under Public Service in Vegetables and Herbs.

The field diagnostic toolbox

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The Field Diagnostic Toolbox is an innovative portable laboratory system designed to revolutionize field data collection and agricultural decision-making. Equipped with a suite of contact and proximal sensing sensors, the toolbox enables rapid, accurate, and in-situ measurement of key physical, chemical, and biological indicators from soil and vegetation. By eliminating the delays and logistical challenges associated with traditional laboratory analysis, it enhances the efficiency and responsiveness of agricultural monitoring. A critical advantage of the toolbox lies in its real-time data acquisition and transmission capabilities. Through a user-friendly Android application compatible with smartphones and tablets, data collected in the field are seamlessly transferred to a centralized GeoDatabase. There, they are integrated with background datasets, including satellite imagery, and undergo debugging, storage, and correlation processing to ensure quality and usability. This Toolbox offers more than rapid diagnostics—it provides a comprehensive decision-support service for farmers, agronomists, and land managers. It supports precision agriculture by delivering optimized, spatiotemporal recommendations for irrigation, fertilization, and crop protection. Real-time remote access allows agronomists to monitor field conditions and offer expert advice without the need for physical presence, enhancing responsiveness and efficiency. The Toolbox supports environmental sustainability and regulatory compliance. It facilitates monitoring of agricultural practices in line with Good Agricultural Practices (GAP) and supports reporting for subsidy programs (e.g., CAP, eco-schemes). Additionally, it enables farmers to document carbon-sequestration activities like no-till farming, cover cropping, and compost application, thus unlocking opportunities in carbon credit markets through MRV (Monitoring, Reporting, and Verification) protocols. Beyond diagnostics, the Toolbox functions as a “digital agronomist,” offering automated alerts, historical data access, and actionable recommendations. It supports the sustainable use of resources and improves long-term farm planning. By bridging real-time field data with expert services, the Field Diagnostic Toolbox transforms conventional agricultural management into a smart, data-driven, and ecologically responsible process.

Keywords: *smart agriculture; real-time field diagnostics; good agricultural practices (GAP); carbon monitoring (MRV); Sustainable farming*

Nutritional quality of field crops: Cereal grains and groats example

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The nutritional value of cereal grains and their minimally processed fractions plays a central role in promoting sustainable and health-conscious food systems. This study summarises the results of recent research on wheat, spelt, barley, oats and buckwheat, focusing on the effects of genotype, field management (organic vs. conventional) and post-harvest processing (threshing, hulling, brushing/polishing) on compositional properties. Significant differences were found between species and varieties in the profiles of protein, fat, fibre, β -glucan and elements. Spelt was consistently higher in protein, while barley and oat groats were rich in β -glucan, a soluble dietary fibre associated with cardiovascular and blood glucose benefits. Organic farming methods increased fibre and β -glucan content in several cases, particularly in spelt and barley. The processing methods influenced the nutritional composition of the groats. Brushing/polishing had a minimal effect on nutrient loss, indicating that they are suitable for the production of ready-to-eat whole-grain products. Elemental analysis by ICP-MS revealed that wholemeal fractions contained higher concentrations of macroelements (K, P, Mg) and microelements (Fe, Mn, Zn) compared to refined flours. In buckwheat, the husks were rich in Cr, Ca and Na and had a low protein and fat content, but were unsuitable for direct consumption. Multivariate analyses (PCA, ANOVA) confirmed that species and processing fractions are the most important discriminators of nutritional quality. The results support the use of customised processing strategies for each cereal species: minimal polishing for wheat and spelt, and more intensive polishing for barley, oats and buckwheat to obtain nutrient-rich groats. This research emphasises the importance of integrating crop science, sustainable farming practices and gentle processing technologies to improve the nutritional profile of cereal-based foods. The results are valuable for breeders, processors and consumers seeking functional and healthy cereal products.

Keywords: *whole grains; groats; nutritional composition; organic farming; grain processing*

Acknowledgment: This study was supported by the research programme Agrobiodiversity (P4-0072; ARIS), the Functional Food for the Future project co-financed by the Republic of Slovenia and the EU under the European Regional Development Fund (GA No. C3330-16-529005), the Public Service in Agriculture co-financed by the Ministry of Agriculture, Forestry and Food of the Republic of Slovenia and the SLO-HUN bilateral project (ARIS No. 5100-4/2018-25 – 2018-2.1.11-TÉT-SI-2018-00010).

Detection of root pests in maize using proximal hyperspectral imaging

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Maize production relies heavily on adequate water availability throughout the entire growth cycle to ensure both high yield and crop quality. Due to its shallow root system, maize is particularly vulnerable to water deficits and soil nutrient limitations. Integrated management strategies aim to address these challenges by reducing pesticide use while maintaining or enhancing yields. Hyperspectral imaging presents a promising non-invasive approach for detecting both biotic and abiotic stress factors in crops. This imaging technique captures detailed reflectance data across wavelengths ranging from 400 to 2500 nm, offering insights into plant pigment composition, leaf cellular structure, and moisture status. In this study, hyperspectral images were analyzed from two maize hybrids, ZP 341 and Futurixx, across several treatment combinations. These included the presence or absence of drought, and two root pests (*Meloidogyne luci* and *Diabrotica v. virgifera* larvae). Images underwent radiometric correction and segmentation, followed by conversion into reflectance values. Pre-processing of mean spectral signatures included detrending, signal noise variates, second-order Savitzky-Golay derivatives, and sparse partial least squares discriminant analysis. An explainable hierarchical classification approach with support vector machines was used. A hierarchical classification approach achieved a detection accuracy (measured by F1) of infected plants between 0.6 and 0.75, depending on variety and watering status. Best results were achieved with well-watered ZP341 plants (F1 = 0.75), and lowest with water-deficient Futurixx (F1 = 0.6). Overall, accuracy was highest with well-watered plants. Variety identification achieved a moderate F1 of 0.7, while drought detection was most reliable in ZP341 (F1 = 0.83; Futurixx F1 = 0.75). These results indicate that hyperspectral imaging enables identification of root pests and other stressors in maize, independent of hybrid type or environmental stress, supporting its potential role in sustainable crop monitoring and management.

Keywords: crop monitoring; biotic and abiotic stress; drought detection; spectral analysis; plant pigment composition

Amplifying the societal impact of European Funded research programmes: how to engage stakeholders and share best practices

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Who can shape the future of Agriculture? The farmers? The researchers? The consumers? All of the above? Surely, all of the above and many more, the so called stakeholders. In every programme or project there are individuals or organisations that are either positively or negatively affected by it. Yet, their perspectives, when taken into consideration, can enrich the researchers point of view and amplify the results and the impact of their work. European funded programmes are aiming to increase their societal impact in order to bridge the gap between research and society and validate that research outcomes are both usable and useful. Designing engagement plans and sharing best practices between research organisations can augment their societal impact effectively.

Keywords: *societal; impact; research; programmes*

Valorization of Italian maize germplasm in the frame of Plant Genetic Resources for Food and Agriculture (RGV-FAO) program

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Italy is characterized by many different climatic and orographic environments, which produced a wide set of maize landraces, promoting selection of flint and semi-flint maize with high milling yield, suitability in food flour production, pasting properties, and wide genetic variability for main grain components. The CREA genebank in Bergamo preserves a large maize collection: over 4,000 accessions, among which 720 Italian landraces and 600 Italian inbred lines, are maintained ex-situ at 5°C and periodically regenerated in field trials under controlled pollinations. In the framework of several research programs, CREA takes care of the conservation, use, and valorization of Italian maize genetic resources; in particular, the activities developed during 2020-2024 in the Plant Genetic Resources for Food and Agriculture (RGV-FAO) Program are illustrated here. Maize Italian accessions (197 landraces and 140 inbred lines) were characterized in field trials by phenological and morphological traits, such as days to tasseling and silking, plant height, ear length and diameter, number of rows and kernels/row, kernel type, kernel color, cob diameter and cob color. Grain chemical composition (protein, lipid, fiber, ash, and starch) was determined on 42 traditional landraces and 8 maize pigmented accessions developed by crossing Italian varieties with Bolivian “Morado” and Mexican “Azul” germplasm. This characterization showed large biodiversity within the CREA maize genebank.

Keywords: *maize; genebank; biodiversity; grain composition*

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Mining maize gene bank diversity: A pillar for future-ready breeding

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The agricultural sector must reconcile environmentally sustainable practices with producing healthier, diversified food and feed while maintaining productivity amid climate change. The Maize Research Institute Zemun Polje (MRIZP) genebank is an invaluable repository of extensive maize genetic diversity. Efficient mining of this resource contributes to uncovering superior genotypes that underpin successful breeding initiatives. Over the past decade, research has demonstrated that pre-breeding effectively connected maize genetic resources with applied breeding programs. During this phase, 31 local landraces—out of 321 identified as promising for drought tolerance—were selected based on their superior agronomic performance per se and further evaluated, considering heterotic patterns, yield, and grain chemical composition. The introgression of landraces with the highest levels of targeted quality parameters per se—protein, starch, and lipids—will enrich elite breeding material and bolster MRIZP’s programs focused on quality improvement. Additionally, 63 early-maturing, predominantly flint-type landraces from the drought-tolerance subset set the foundation of a novel pre-breeding initiative aimed at broadening the genetic base of early-maturing elite maize varieties. Furthermore, 572 MRIZP genebank inbreds, characterized by collection-site attributes from Southeast Europe (the MRIZP-SEE sub-panel), were genotyped using the Axiom 600k Array. Admixture analysis revealed that 455 of these inbreds clustered into distinct populations—A374, European Flint, and Wf9—highlighting clear genetic differentiation. Signatures of selection on several chromosomes highlighted stress response genes associated with climate adaptation. Moreover, the MRIZP genebank has contributed 96 maize landraces to the European Evaluation Network for Maize (EVA), part of the European Cooperative Program for Plant Genetic Resources (ECPGR). EVA provides standardized phenotypic data from multi-location trials across Europe, assessing traits relevant to breeders, abiotic and biotic stress resistance, as well as genotyping information. Through strategic public-private partnerships, this comprehensive dataset helps identify superior accessions for breeding to support climate resilience, sustainability, food security, and crop improvement.

Keywords: *genotyping; heterotic pattern; phenotyping, pre-breeding; Zea mays L.*

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Mining Allelic diversity of maize landraces for abiotic and biotic stresses

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Landraces are valuable sources of genetic diversity for addressing the challenges of climate change, reducing fertilizers and pesticides but remain underutilized in modern breeding programs. To fill this gap, the MineLandDiv project (Mining Allelic Diversity in Landraces for Tolerance to Abiotic and Biotic stress) aims at (i) identifying maize landraces and favorable alleles for tolerance to abiotic (heat/drought - cold - nitrogen) and biotic stresses (Corn borer) that could be used to broaden genetic diversity of modern breeding germplasms and (ii) better understanding their resilience to variable environmental conditions. We optimized a panel of 341 temperate maize landraces representative of European genetic diversity that we evaluated for roots architecture in platform and for various agronomic and physiological traits across a field trial European network of 6 locations using high throughput phenotyping / envirotyping tools. We observed a strong effect of stress for yield according to treatments and environments. We genotyped DNA pools of these landraces with 600 000 SNPs to conduct genome wide association studies with traits and environmental variables from landrace collecting sites. We identified genomic regions involved in trait and environmental variation. We developed a DNA pool targeted sequencing approach to compare haplotype contents at 14 genomic regions involved in tolerance to abiotic stresses using 372 landraces and 342 inbred lines. We showed that 17% of genetic diversity found in landraces has been lost in inbred lines. We developed genomic prediction that predicted with high accuracy agronomic traits in different environments. First results are encouraging to identify promising alleles and landraces for prebreeding.

Key words: genetic resources; genebanks; prebreeding

Genetic structure and diversity of maize lines from the MRIZP gene bank pre-breeding program

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The use of genetic resources in maize breeding is crucial for both conserving biodiversity and harnessing desirable genetic variability. Maize breeding depends on understanding heterotic patterns and effectively exploiting heterosis. However, limited information on the genetic structure and heterotic patterns of gene bank materials often hampers efforts to enhance or integrate these resources into breeding programs. In this study, we selected 87 lines from the Maize Research Institute Zemun Polje (MRIZP) pre-breeding program to analyze their genetic structure in relation to seven lines with known pedigrees, using 25k SNP markers. Among these, 15 lines are part of the MRIZP gene bank mini-core collection for drought tolerance. Twenty lines were developed through pedigree selection from crosses between local landraces and three commercial testers (T1 - Iowadent, T2 - BSSxIowadent, T3 - Lancaster). The remaining 52 lines were derived via pedigree selection from three synthetics (BS22, N425, and IP3712) and three local landraces (K1952, K1492, and Bosanac) with unknown genetic backgrounds. Analysis revealed that lines from BS22, N425, K1952, and Bosanac formed distinct clusters that did not include commercial lines. Fourteen drought-tolerant mini-core lines, along with IP3712 and lines resulting from crosses between local landraces and T2, clustered together with lines T1, B73, A632, and CA12053. Conversely, T3 and its derived lines formed a separate cluster. Lines originating from Bosanac contained germplasm not present in any of the other tested lines, with only minor contributions from Lancaster germplasm. Lines resulting from crosses between local landraces and T1 clustered with ex-PVP lines and one drought-tolerant mini-core line (Ku 52L-5g7379). These results suggest that introgression of new variability present in these lines—many of which already demonstrate good performance per se—can significantly support sustained progress in maize breeding efforts.

Keywords: *genetic resources; genetic background; SNP markers; Zea mays L.*

Acknowledgment: This study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (Grant No. 451-03-136/2025-03/200040).

Per se and test cross performance of Croatian maize landraces for grain yield

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Today, maize cultivation is dominated by hybrids, which have almost completely replaced the landraces and thus contributed to the reduction of genetic diversity. The maize landraces stored in the world's gene banks represent a potential resource of currently unutilised indigenous diversity that could be harnessed for crop improvement. To evaluate the breeding potential of Croatian maize landraces, 60 test crosses were produced by crossing 30 landraces with two inbred testers. During the 2024 growing season, two field trials were sown side by side at three locations (Šašincevec, Rugvica and Osijek), one with 30 landraces and two testers and the other with 60 test crosses and five commercial hybrid checks. Combined ANOVA across locations revealed a significant effect of genotype (G), location (E) and G × E interaction for grain yield (GY) in both trials. Analysis of combining ability for GY across locations showed that general combining ability with respect to landrace and tester contributed 50 and 8%, respectively, to the total sum of squares of the test cross, while the relative contribution of specific combining ability was 42%. The average GY of the 30 landraces per se at Šašincevec, Rugvica, Osijek and across locations was 7, 4.4, 2.3 and 4.6 t/ha respectively, while the corresponding average GY of the test crosses was 12.2, 6.9, 4.4 and 7.7 t/ha respectively. However, a large variation in GY was observed among landraces and their testcrosses with both inbred testers, resulting in a wide range of estimated better parent heterosis for GY (performance of the testcrosses relative to their landrace parents), which varied from 118 to 277%. Of the 60 test crosses, 17 test crosses achieved GY at the level of the average of five commercial hybrids, while seven of them were at the level of the highest-yielding hybrid.

Keywords: maize; landrace; hybrid; grain yield; combining ability

Predicting adaptation of maize landraces from genebanks by combining DNA-pool genotyping, genomic prediction and genomic offset

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Due to their local adaptation to contrasted environments, maize landraces are a valuable source of genetic diversity for facing climate change and low input agriculture challenges, however they are underutilized and undercharacterized. High-throughput pool genotyping (HPG) has proven to be efficient to characterize genetic structure, gene diversity (Hs) and identify selective sweeps in landraces. Here, we investigated the interest of genomic prediction (GP) and genomic offset (GO) approaches using HPG to predict adaptation of landraces to various environments. To do so, 397 European landraces were evaluated for yield, plant height, and flowering time in the ECPGR multi-environment trials of the European Evaluation Network (EVA) network including 25 environments. The objectives were to : i) evaluate the effect of Hs and GO on yield, height and flowering, ii) predict these traits using GP under different cross-validation (CV) scenarios, and iii) evaluate the effect of including Hs and GO in the GP model. Gene-diversity of landraces (Hs) was positively correlated with yield ($R^2 = 0.29$) and plant height ($R^2 = 0.28$) while GO was negatively correlated with these two traits. GP yielded high predictive abilities that varied according to cross-validation scenario, reaching a minimum of 0.52 when predicting new genotypes in new environments. Integrating Hs and GO in the GP model increased predictive abilities by up to 13.2% when predicting new genotypes in a new environment. Our results show that GP using HPG is a promising tool to predict the value of maize landraces in genebanks, notably their adaptation to various environments, and that including gene diversity and genomic offset in the prediction model could improve predictive abilities.

Keywords: bulk genotyping; genomic prediction; genomic offset

Genetic resources and agrobiodiversity: Foundations of sustainable agriculture in Serbia

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In addition to wild biological species, domesticated species also represent an important component of species diversity, forming agrobiodiversity as an integral part of overall biodiversity. Agrobiodiversity encompasses all microorganisms, plants, and animals essential for maintaining the structure and function of agroecosystems, as well as for ensuring the sustainable production of safe and healthy food. Also, agrobiodiversity, is the central object of studying agroecology. The aim of this paper is to point out, based on the available scientific literature, the importance of agrobiodiversity in preserving ecosystem sustainability and food production. The research presented in this manuscript is theoretical and of an overview nature, based on data analysis of information collected from relevant scientific journals. A review of the literature, including the scientific works of the author of this paper, indicates that a significant part of the total species diversity of Serbia is represented by cultivated plant and animal species that constitute a significant genetic resource. Most varieties are the product of the selection of suitable species and artificial selection. 193 plant species are grown in Serbia, which include cereals, industrial and fodder plants, vegetable plants, fruits, and vines, and constitute a significant genetic diversity. It is estimated that around 25. 000 seed accessions of old varieties and local populations are stored in agricultural institutions in Serbia today. Most of the plant species grown in Serbia have their wild relatives that grow wild in natural ecosystems. Based on the review of conducted scientific research, it can be concluded that the preservation of agrobiodiversity is a key factor that enables the sustainability of the ecosystem, the survival of living organisms on Earth, as well as the production of safe food.

Keywords: *agrobiodiversity; biological species; microorganisms; plants; animals*

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Research trends in maize breeding: A global perspective from bibliometric and topic modeling approaches

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Maize breeding has undergone significant changes over the past two decades, primarily driven by the rapid development of new technologies, particularly artificial intelligence. Alongside genomic selection, which has already been implemented in commercial breeding programs, highly sophisticated tools for prediction, decision-making, data integration, and interpretation now form the foundation of the modern approach to maize breeding. To understand the impact of latest technologies on maize breeding, we conducted a comprehensive literature analysis by applying Bibliometric science mapping and Latent Dirichlet Allocation (LDA). We formulate the following search query to retrieve the publications from the Web of Science Core Collection: ALL: ("artificial intelligence" OR "machine learning" OR "deep learning" OR "neural network" OR "AI" OR "ML" OR "genomic prediction") AND ALL: ("maize" OR "corn") AND ALL: ("breeding" OR "genetic improvement" OR "selection"). After the detailed article selection and data preparation, 480 publications of 1.643 search results were used as the input for the analysis. The overall trend of the advanced maize breeding literature showed that the number of publications increased during the observed time-span (2010-2024) with the annual growth rate of 32.8%. In total, 60 countries are active in the field of advanced maize breeding with the high degree of the international collaboration. The top five countries according to the number of publications were: China, USA, Mexico, Germany and Brazil. Latent Dirichlet Allocation (LDA) analysis revealed fifteen topics, indicating three main domains influencing advanced maize breeding; i) genetics and genomics; ii) computer science and machine learning; and iii) phenotyping and sensing. The analysis provides important information regarding the future direction of maize breeding and highlights anticipated developments associated with the integration of novel technological advancements.

Keywords: *Web of Science; bibliometrics; latent dirichlet allocation*

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Optimizing maize yield prediction using Gradient Boosting Machine with correlation-based feature selection

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Accurate yield prediction in maize is essential for improving decision-making in agronomic management. This study presents a machine learning pipeline combining outlier detection, correlation-based predictor filtering, and Gradient Boosting Machine (GBM) modeling. The field experiment was conducted in 2024 at the Rinski Šančevi research station (Novi Sad, Serbia) using a split-plot design. Four plant densities served as main plots, while six maize hybrids were arranged as subplots. Yield and a set of spectral and canopy-related variables were collected from the experiment, via combine harvester and UAV DJI Mavic 3E. The dataset was split into training (70%) and testing (30%) subsets. Outliers in the training set were assessed using the interquartile range (IQR) method, with no values exceeding the $\pm 3 \times \text{IQR}$ thresholds. Correlation analysis was applied to select variables with an absolute Pearson correlation coefficient ≥ 0.40 with yield, resulting in the retention of nine predictors: Plant Height, Color Index of Vegetation Extraction, Excess Green minus Excess Red, Excess Red Index, Mean Green-Red Vegetation Index, Normalized Green-Red Difference, Visible Atmospherically Resistant Index, Visible Band Difference Vegetation Index and Percent Vegetation Cover Area. A GBM was trained using repeated 10-fold cross-validation (5 repeats) with hyperparameter tuning. The best-performing model used 500 trees, a learning rate of 0.01, interaction depth of 1, and a minimum of 5 observations per node. On the training set, the model achieved an RMSE of 613.3 and $R^2 = 0.66$. On the test set, RMSE was 762.3 and $R^2 = 0.744$, indicating strong generalization. These results demonstrate that vegetation indices derived solely from visible light bands — particularly those emphasizing green-red contrast and canopy structure — can serve as effective predictors of maize yield. The integration of basic filtering procedures with a robust ensemble model enabled accurate prediction from RGB-based phenotyping data in a multi-factor field trial.

Keywords: *maize; UAV phenotyping; visible-band vegetation indices; machine learning, canopy trait analysis*

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The role of combining abilities in maize inbred lines for developing high-yielding hybrids

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Combining abilities refers to the performance of parental lines in hybrid combinations and is a crucial concept in maize breeding. It provides insight into the genetic potential of inbred lines and helps breeders identify superior parents for hybrid development. Effective combiners are not only genotypes with high combining abilities but must also exhibit other desirable traits. Combining abilities are typically assessed using diallel, line x tester mating design, followed by analysis of variance to estimate general (GCA) and specific (SCA) effects. The results guide breeders in selecting the most promising parental lines and heterotic groups, ultimately leading to the development of high-yielding and stable maize hybrids. The aim of this study was to evaluate whether inbred lines with the highest absolute general combining ability values, whether positive or negative, produce hybrids with the highest specific combining ability values for grain yield and its components. Seven maize inbred lines were crossed using a diallel method to generate 21 hybrids, and the experiment was conducted in two years at three locations. A completely randomized block design with three replications was employed for both inbred lines and hybrids. Inbred lines ZPL2 and ZPL4 exhibited the highest general combining ability values for grain yield, ZPL2 and ZPL7 for ear length, ZPL4 and ZPL5 for number of kernel rows, and ZPL2 and ZPL7 for number of kernels per row. Hybrid combinations of ZPL1 x ZPL4 and ZPL2 x ZPL4 showed the highest specific combining ability values for grain yield, ZPL1 x ZPL4 and ZPL2 x ZPL6 for ear length, ZPL1 x ZPL4 and ZPL5 x ZPL6 for number of kernel rows, and ZPL1 x ZPL4 and ZPL2 x ZPL6 for number of kernels per row. Based on the results for each trait, it is clear that it is sufficient for one parent to have high general combining ability values in order to achieve high specific combining ability values when crossed with another parent.

Keywords: combining abilities; maize; grain yield; yield components

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Evaluation GEM Project accessions as potential sources for broadening genetic basis of temperate breeding programs

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Forty four accessions obtained from Germplasm Enhancement of Maize (GEM) project, an American pre-breeding program were examined, 20 belonging to the Non-Stiff Stalk (NSS) heterotic group (GEMN) and 24 accessions classified in the Stiff Stalk (SS) heterotic group (GEMS). Accessions were crossed with two corresponding testers from the opposite heterotic group, namely GEMN accessions with ex-PVP BSSS line PHHB9 and Novi Sad commercial BSSS 3616NS line, and GEMS accessions with ex-PVP NSS inbred line PHN82 and Novi Sad commercial NSS line 66791NS. In addition to the 85 test-crosses, seven commercial F1 hybrids were included in the field trials as checks (FAO 600 maturity group). The field trials for grain yield and moisture content were conducted at four locations according to the incomplete block design. Five test-crosses were identified as promising, due to grain yield that exceeds most of commercial check hybrids. All perspective test cross hybrids were obtained by crossing the BSSS tester with NSS GEM materials and the vast majority (four out of five) were obtained with the PHHB9. In addition to the grain yield, grain moisture content at harvest is also an extremely important trait. As expected, the majority selected hybrids based on the yield had higher grain moisture content at harvest than the commercial checks. However, hybrid No. 58, which achieved grain moisture content at check hybrid level with a grain yield that exceeds most of the check hybrids in the trial, stands out. As potential sources of desirable grain yield alleles for the improvement of NSS temperate materials and broadening of the genetic basis, five selected accessions can be used, and especially the accession GEMN-0246 that participated in the No. 58 hybrid, which achieved a respectable yield with moisture content on the level of the FAO 600 maturity group.

Keywords: *GEM lines; exotic germplasm; grain yield; grain moisture; pre-breeding; maize*

Acknowledgment: Maize Department, Institute of Field and Vegetable Crops, Novi Sad.

The analysis of maize inbred lines of various cycles of selection

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The evaluation of combining abilities is an important stage in the development of high-yielding maize hybrids. This study was focused on inbred lines derived from the populations BSSS and BSCB1. The aim of the study was to observe combining abilities, heterosis and variability of inbred lines and their hybrids. According to the analysis of results gained on the number of kernels per row, highly significant positive values of general combining ability (GCA) and specific combining ability (SCA) were observed at the locations Zemun Polje and Kraljevci during the two-year investigation. The dominant gene action prevailing in inheritance was established in inbreds derived from synthetic BSSS and in the parental inbreds of the elite hybrid. This was shown by the GCA to SCA ratio that was less than unit. Additive variance was greater than non-additive one in inbreds derived from the synthetic BSCB1 and inbreds of the elite hybrid. The highest, i.e. the lowest average number of kernels per row was determined in the hybrid ZPL2 x B73, i.e. the hybrid B90 x B91, respectively. At the same time, the highest heterosis was established in the hybrid B90 x B91. The inbred B73(C5) derived from the synthetic BSSS was the best general combiner, which shows that it can be used in breeding programmes designed to increase the number of kernels per row. The inbred B97(C9) derived from the synthetic population BSCB1 expressed high GCA values.

Keywords: *maize; combining ability; number of kernels per row; heterosis*

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Reciprocal effect on grain yield and agronomic traits in single-cross maize hybrids

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Reciprocal effect in maize refers to the phenotypic difference between reciprocal F_1 hybrids. The aim of this study was to estimate the influence of reciprocal crosses on grain yield, grain moisture percentage at harvest, plant height, primary ear height, and the number of leaves above the primary ear. Five single-cross hybrids, their reciprocal crosses and six parental inbred lines were selected. All hybrids belonged to the late maturity group (FAO 500-600), with the Lancaster ZPL-7 line as a common paternal parent, i.e., as the maternal parent in their reciprocal crosses. The experiment was set up in two replications using a completely-randomized block design in 2016 and 2017, at a total of seven environments. A three-factorial ANOVA was performed, with hybrid, reciprocal effect and location as factors. The reciprocal effect significantly influenced grain yield, grain moisture, plant height and ear height, while hybrid and location were significant for all examined traits. Considering all five hybrids individually, one hybrid displayed a significant reciprocal effect for grain yield, one for ear height, and two hybrids for grain moisture and plant height. The effects were both positive and negative, depending on the genotype itself. No significant differences were observed for the number of leaves above the primary ear between normal and reciprocal variants. The largest difference in grain yield was recorded between ZP 606 reciprocal (12.06 t/ha) and ZP 606 normal (11.28 t/ha). Pearson correlations were calculated among the examined traits. All correlations between grain yield and other traits were significant and positive. The reciprocal effect has a strong influence on the measured traits, but it is genotype-specific. Therefore, in future studies, normal and reciprocal variants of all commercial maize hybrids should be examined.

Keywords: grain yield; morphological traits; reciprocal effect; single-cross; *Zea mays* L.

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Grain yield and grain yield stability of pre-commercial and commercial ZP maize hybrids (FAO 400, 500 and 600 maturity groups)

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Every year, Maize Research Institute Zemun Polje conducts multi-environment strip field trials with pre-commercial, recently released and currently best-selling ZP maize hybrids, to evaluate their grain yield performance and stability. In 2024, ten ZP hybrids (FAO 400-600 maturity groups) were selected for these trials, out of which three were pre-commercial (candidates for commercialization), four were recently released, and three currently best-selling used as checks. The trials were conducted at 40 sites across all main maize growing regions in Serbia, and 26 sites were chosen as appropriate for taking into consideration. The plot area was approximately 1000 m². The highest average grain yields were recorded in three recently released hybrids ZP 5550 (7661 kg ha⁻¹, 1st highest), ZP 5601 (7557 kg ha⁻¹, 3rd highest) and ZP 4790 (7488 kg ha⁻¹, 4th highest), and in pre-commercial hybrid ZP 4722 (7588 kg ha⁻¹, 2nd highest). These results confirmed the high performance of recently released hybrids, while hybrid ZP 4722 is proposed for commercialization consideration. The grain yield stability analysis was performed by using the linear regression model (Eberhart and Russell, 1966) in order to reveal important information about hybrid performance pattern across different environments/growing conditions. The hybrids ZP 4242 and ZP 4722 exhibited good response to good growing conditions (regression coefficients b_i were 1.14 and 1.09, respectively) which makes them suitable for the best farmers and the best growing practices. The hybrids ZP 5601 and ZP 6263 showed good adaptation to poor growing conditions (b_i were 0.88 and 0.93, respectively), while hybrids (ZP 457, ZP 4744, ZP 4790, ZP 5011 and ZP 606) with b_i values near 1 showed stable performance across all environments. The highest yielding hybrid ZP 5550 showed moderate adaptation to poor growing conditions ($b_i=0.95$), and, as such, it is highly recommended for Serbia's growing conditions, which are becoming less favorable due to frequent droughts. For more reliable conclusions, multi-year data should be taken into consideration.

Keywords: *maize hybrids, multi-environment trials, grain yield, yield stability, recommendations for farmers*

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The non-parametric estimation of 1000-seed weight stability of maize inbred lines

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The present study encompassed the estimation of 1000-seed weight stability of 12 maize inbred lines with three different types of cytoplasm: cytoplasmic male sterility - type C (cms-C), type S (cms-S) and fertile cytoplasm. The trials were carried out at two locations in Zemun (Selekciono polje and Školsko dobro) in 2020 and 2021 according to a randomised complete block design with three replications for each type of cytoplasm. The 1000-seed weight ranged from 277.8 to 368.0 g, 264.5 to 349.8 g and from 259.8 to 373.3 g in inbreds with C, S cytoplasm and fertile cytoplasm, respectively. Stability parameters (Si(1) - mean rank difference and Si(2) - rank variance) were used to estimate the stability of genotypes, while tests of significance Si(1) and Si(2) indicated statistically significant differences in the 1000-seed weight among genotypes. The inbreds L10 and L7 were the most and the least stable, respectively, within the group with C cytoplasm. In relation to inbreds with S cytoplasm, L10 was also the most stable, while L12 was the least stable. Moreover, the inbred L10 was also the most stable among the inbreds with fertile cytoplasm, while the inbred L6 was the least stable. The graphic analysis showed different stability intervals among groups, while the critical values of the χ^2 test confirmed the validity of the applied non-parametric methods. The results emphasise the importance of using non-parametric measures of stability for the precise assessment of adaptability and consistency of seed weight in various maize genotypes, which is important for breeding and selection and for a further development of stable inbred lines under changeable agro-ecological conditions.

Keywords: 1000-seed weight; inbred lines; cms; stability of genotypes

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Effect of growing season rainfall on maize productivity in the region of Knezha

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The Knezha region, located in Bulgaria, is a traditional producer of maize for grain under non-irrigated conditions. However, yields have shown considerable interannual variability, primarily driven by fluctuations in hydrothermal regimes. Both local and regional changes in climatic and agro-climatic parameters indicated an increase in air temperature and abiotic stress during the growing season. There wasn't any clear tendency of change in precipitation, but with an established change in annual distribution. This study investigated the relationship between wetting conditions and grain yield performance of maize hybrids classified in FAO maturity group 500. The analysis was based on 30 years of yield data (1991–2020) collected from field trials conducted at the Maize Research Institute in Knezha, where all hybrids were grown under uniform agronomic practices to minimize management-related variation. Agrometeorological characterization focused on three key indices: cumulative precipitation, potential evapotranspiration (ET_r), and the balance of atmospheric humidity (BAO), particularly during the period encompassing leaf development and reproductive stages, which commence in the final third of June for this hybrid group. To evaluate the influence of these parameters, principal component analysis (PCA) was applied as a multivariate statistical approach capable of identifying the most explanatory climatic drivers. The results of PCA indicated that the most significant contributors to grain yield variability were the total precipitation and the atmospheric humidity balance recorded during the generative stage. The analysis showed that the sum of rainfall and balance of atmospheric humidity during the generative stage has the greatest effect on grain yields of maize FAO group 500. The study emphasizes the need to account for agrometeorological indices in hybrid evaluation protocols and yield prediction models, particularly in the context of increasing climatic instability and the intensification of heat and drought stress.

Keywords: *agro-climatic conditions; rainfall sum; hydrothermal index; mid-late maize hybrids*

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The influence of the production method of different sweet corn hybrids on the productive characteristics

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Sweet corn (*Zea mays* L. var. *saccharata* Sturt.) is an important vegetable crop due to its nutritional value and economic profitability. The work aimed to examine the influence of the production method (container and direct sowing) on the productive characteristics (ear length, grain depth and ear weight) of two supersweet sweet corn hybrids (DDST 255 and Sweet Nugget) under irrigation conditions. The experiment was conducted in 2024 on a family farm in Bogojevac, southern Serbia (43.0538316, 21.9677546, 225 m above sea level) in an open field. Land preparation included fall ploughing and spring pre-sowing preparation, along with the installation of a drip irrigation system. Sowing and transplanting were done on May 18, 2024, with a distance between strips of 70 cm, a row spacing of 50 cm and a distance between plants of 20 cm. Harvesting was done on July 10 (container seedling production) and July 27 (direct sowing). Data were analysed using ANOVA and the LSD test ($p < 0.05$ and $p < 0.01$) in IBM SPSS Statistics 26.0. Container production resulted in greater ear length (DDST 255: 22.50 cm; Sweet Nugget: 17.70 cm), kernel depth (DDST 255: 1.22 cm; Sweet Nugget: 1.19 cm) and ear weight (DSST 255: 220.00 g; Sweet Nugget: 177.90 g) compared to direct seeding. The method of production had a statistically significant influence ($p < 0.01$) on all investigated traits. In contrast, the interaction of the method of production and the hybrid was significant only for the weight of the ear. Hybrid DDST 255 showed higher values compared to Sweet Nugget. Container production significantly affected the productive characteristics of sweet corn compared to direct sowing, especially in hybrid DDST 255. The choice of production method should be adapted to the choice of hybrid in order to achieve satisfactory values of the most important productive characteristics of sweet corn.

Keywords: *sweet corn; container production; direct sowing; productive traits*

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The influence of locality on the agronomic characteristics of maize hybrids

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During 2024, maize trials were set up at two locations: Rača Kragujevacka and Sombor with two maize hybrids: H1-BC 306 and H2-BC Klipan, in four replications. The influence of hybrids and locality on the morphological characteristics and components of maize a thousand grain mass was examined. The following parameters were tested: maize plant height, cob length and 1000 grain mass. Genotype and locality had a statistically significant effect on the 1000 grain mass, plant height and maize cob length. The average 1000 grain mass in both locations was 317.5 g for the H1 maize hybrid and 387.5 g for the H2 hybrid. The results showed that higher values of the tested maize parameters were recorded in hybrid H2 compared to hybrid H1. At the Sombor location, higher values of the tested hybrids for the 1000 grain mass were recorded compared to the Rača Kragujevacka location, while at the Rača Kragujevacka location, higher values were recorded for plant height and maize cob length for hybrid H1 (219.5 cm; 19.65 cm) and hybrid H2 (228.1 cm; 24.45 cm). Based on the obtained results, it is evident that both maize hybrids achieved satisfactory results in both localities.

Keywords: *plant height; cob length; 1000 grain mass; Zea mays L.*

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SoyPredict: Data-Driven strategies of soybean breeding

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The goal of SoyPredict is to increase the efficiency of breeding new soybean varieties that are suited to the agro-climatic conditions of Serbia and Europe by using sophisticated breeding tools, multi-omics data, and mathematical modeling for yield prediction. The project focuses on soybean, a commodity of significant economic and environmental significance, by creating long-term breeding plans utilizing cutting-edge technologies. The main consequences of climate change in Serbia are reflected in the reduction of the yield of the main crops. Taking into account the negative effects of climate change on agricultural production in general, the availability of new and improved soybean varieties is an important component in overcoming the negative effects. SoyPredict ambition is the development of an efficient, long-term breeding strategy by combining different prediction models based on big data of different aspects of soybean biology. The term ‘breeding strategy’ implies a plan to optimize the production of soybean varieties using the full suite of technologies and to release the improved varieties faster. Specific strategies will determine how to integrate traditional and multi-omics approaches for improving the efficiency of a breeding process. The primary idea behind SoyPredict is the ability and accuracy of GP, HTPP, and PP models to predict soybean yield (both within and between environments, at various stages of genotype evaluations), as well as the implementation of model compilation and the development of cost-effective strategies for the application of one or more models. SoyPredict’s goal is to establish the foundation for economically viable soybean breeding so that improved varieties can be continuously released to meet the demands of industry, producers, and consumers. Improved breeding due to access to high-tech breeding tools and methods, will reside on mechanisms for long-term crop improvement and overcoming the negative impacts of climate change.

Keywords: *soybean; breeding; prediction models*

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Genotype $\times$ environment interaction for grain protein and oil content in mid-season soybean genotypes

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The study aimed to analyse the impact of genotype $\times$ environment ($G \times E$) interactions on grain protein and oil content in soybean genotypes from the Maize Research Institute Zemun Polje collection, in order to identify stable sources for breeding purposes. Sixteen soybean genotypes from maturity group I (mid-season varieties) were examined in field trials conducted over two years at two locations (Zemun Polje and Pančevo), using a completely randomised block design. After harvest, grain protein and oil content were tested using an NIRT (near infrared transmission) analyser. The $G \times E$ interaction for seed protein and oil content was evaluated by the AMMI-1 (additive main effects and multiplicative interaction) model. The first interaction axis of the AMMI-1 model explained 76.6% of the variation in grain protein content in soybean genotypes caused by the $G \times E$ interaction. Six genotypes were positioned close to the stability line, which indicated their greater degree of adaptation to environmental changes. Among these, variety Krizia had the greatest breeding potential due to its higher protein value and good stability. Other genotypes with above-average protein content deviated significantly from the stability line, largely varying in four environments and likely expressing a good adaptation in a smaller number of specific sites. For the oil content, AMMI-1 biplot displayed that the first interaction component accounted for 70.6% of the variation due to $G \times E$ interaction. The largest number of genotypes had a trait value above the general average, differing for the interaction component. Varieties with below average (Laura) and above average oil content (A 1937) showed the greatest stability. Genotypes that produced more oil in grain and did not significantly change their response to environmental changes could be potential sources of germplasm for breeding for increased oil content (A 1937, Hodgson 78, and Shine).

Keywords: soybean; protein and oil content; stability; AMMI-1

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Diurnal variability in chlorophyll fluorescence of soybean under field conditions: Implications for heat stress response

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Monitoring variations in photosynthetic efficiency throughout the day allows the variability of plant stress responses to be studied, helping to improve precision breeding and management. Chlorophyll *a* fluorescence (ChlF) was measured twice in the field during the flowering phase in nine soybean genotypes. The measurements were taken in the morning, which corresponds to optimal environmental conditions, and at midday, which corresponds to elevated temperatures that normally cause stress in the plants. Treatment, genotype and their interaction were highly significant sources of variability ($p < 0.001$) for all ChlF parameters tested. Genotypes differed significantly ($p < 0.05$) in their stress response, expressed as a percentage change in values from morning to midday. The largest average diurnal change was observed for the performance index on an absorption basis (PIabs, 63.8%) and the smallest for the electron transport flux per active reaction centre (ET0/RC, 6.5%). According to the results, heat stress affected both the efficiency with which the absorbed light is used in photochemistry (Fv/Fm) and the electron transport beyond the primary quinone acceptor (ET0/TR0). This was confirmed by the results of the energy fluxes per reaction centre, where ABS/RC, TR0/RC and DI0/RC increased as a result of the high midday temperatures, while ET0/RC decreased. This indicates that although the apparent energy input per active reaction centre increased, less energy was directed into productive photochemistry – that is, the overall photochemical efficiency was reduced. The average PIabs decrease confirms that the overall photosynthetic performance of PSII was impaired and less efficient at high temperatures, indicating a reduced capacity for photochemical conversion. Among the genotypes, photosynthetic efficiency was least impaired in line OS-922 and most impaired in OS-1522. The photosynthetic apparatus of line OS-1322 proved to be very stable under elevated heat, with PIabs remaining almost unchanged as a measure of photosynthetic performance compared to optimal conditions.

Keywords: *chlorophyll a fluorescence; temperature; stress tolerance; breeding*

Response of grain yield and plant traits to ammonium nitrate fertilization in selected *Triticum* genotypes

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The genus *Triticum* encompasses a wide genetic diversity, including both ancient and modern wheat species that are important for agriculture and food security. This study aimed to evaluate the effect of ammonium nitrate (AN) fertilization on grain yield, plant height, and spike length across 23 genotypes of *Triticum* including: spelt (*T. spelta*), durum (*T. durum*), einkorn (*T. monococcum*), emmer (*T. dicoccum*), compactum (*T. compactum*), sphaerococcum (*T. sphaerococcum*), and triticale, under field conditions characterized by low rainfall. The experiment was conducted over two growing seasons (2021–2023) at the Maize Research Institute “Zemun Polje,” Serbia, using a randomized complete block design with two replications. Treatments consisted of a control and fertilization with 150 kg/ha AN applied on March 15. Except for triticale, where yield under AN- fertilized plots was 1% higher, all species showed higher grain yield in control plots compared to AN-fertilized plots, with reductions ranging from 0.32% (durum) to 88.8% (einkorn). This decline is attributed to the dry environmental conditions during the growing seasons, which likely limited water availability and reduced water use efficiency under increased nitrogen supply. Statistically significant differences ($P < 0.05$) between treatments were observed for grain yield in spelt and einkorn, as well as plant height in durum and spelt genotypes. Notably, ancient wheat genotypes such as einkorn and emmer showed greater sensitivity to nitrogen fertilization under dry conditions, likely due to their evolutionary adaptation to low-input environments. Spike length differences were not significant across treatments. The ANOVA revealed highly significant main effects of genotype, year, and treatment, as well as their two- and three-way interactions ($P < 0.05$). The significant Genotype $\times$ Treatment interaction confirms that genotypes responded differently to fertilization. Post-hoc Tukey’s test further grouped genotypes based on yield performance: triticale breeding line ZP 34-15 and durum variety Agaton showed the highest mean yield and were significantly different from the majority of others, while four spelt lines were in the lowest-yield group. These results emphasize the need for integrating genetic diversity with site-specific fertilization strategies to improve wheat productivity and resilience under changing climatic conditions.

Keywords: *Triticum* spp; Nitrogen; Grain yield; Drought stress; Ancient wheat species

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Variation of mass of plant in bread wheat varieties

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The mass of the plant is related to other components involved in forming the yield of wheat. The aim of this study was to estimate the variability of the mass of the plant in 10 genetically divergent wheat varieties. The experiment was set up as a randomized block design with three replications during two year of experiment (2015/16 and 2016/17). The seeds were sown at 0.10 m distances in 1.0 m long rows spaced apart by 0.2 m. Sixty plants at the full maturity stage (20 plants per replication) were harvested and used for analyzing the mass of the plant. The analysis of variance was performed using MSTAT C (version 5.0). Similarities among wheat varieties were analyzed using the hierarchical method of Euclidean distance. The results showed significant differences in average values of the mass of the plant among the analyzed wheat varieties during each year of the experiment. In the first year of the study, the least mass of the plant was recorded for the Morava variety (27.43 g), while the highest mass of the plant was found in the Kremna variety (41.47 g). In the second year, mass of the plant varied from the lowest in the Kosmajka variety (28.77 g), to the highest mass of the plant in the Ljubičevka variety (45.53 g). On average, for all 10 wheat varieties, the mass of the spike was 34.5 g in the first vegetation season and 38.3 g in the second vegetation season, with average value of 36.4 g for all varieties and two years. The highest impact on expression these traits had interaction of genotype/environment (48%) and the least impact had genotype (9%). The differences in the average mass of the plant were determined by genetic and environmental factors, as well as by genotype/environment interaction.

Keywords: *wheat; mass of plant; variety, similarity*

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Biomass quality of the faba bean lines

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Many studies have shown that various parts of the faba bean plant (*Vicia faba* L.) contain useful compounds, including protein, fiber, and a wide range of bioactive substances. The chemical composition of faba bean plant sections varies depending on the plant's genotype and growing conditions. Aside from the grain, the faba bean plant contains relatively little protein. This study evaluated ten promising faba bean lines with high grain protein content identified in prior research. These lines are derived from local Serbian faba bean populations, and previous research indicated that their grain has high protein content (more than 27%). A two-year experiment with two replications was conducted in the experimental field of the Institute for Forage Crops Kruševac, utilizing a random block design. The plants were cut at the green pod stage when the seeds had formed but the plants remained green because higher protein content and higher biomass quality were expected. The following parameters were assessed using the Weende method: crude protein, crude cellulose, ash, crude fat, and nitrogen-free extract (NFE). The results showed significant variance across the studied faba bean lines. Dry mass content ranged from 90% to 91.4%, crude protein content from 18.3% to 23.3%, crude cellulose from 14.2% to 25.6%, crude ash from 6.3% to 9.4%, crude fat from 0.4% to 2.1%, and NFE content from 41.2% to 50.3%. As expected, the protein level in the biomass was high at this stage of mowing, possibly due to the developed grains. This biomass is a high-quality feed for fresh and ensiling applications. Furthermore, the high levels of some of the other assessed parameters indicate that more research is needed on the dry matter content of the entire faba bean plant at this cutting stage.

Keywords: *whole plant; faba bean; mowing stage; feed quality; protein*

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Seed separation of cytoplasmic male sterile and fertile maize lines by NIR (Near Infrared) spectroscopy coupling with chemometric techniques

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Cytoplasmic male sterility (CMS) systems play a crucial role in seed production of hybrid maize. In this system, ensuring the genetic purity of fertile and restorer lines is of strategic importance in hybrid maize production. This necessitates fast and accurate classification methods, among which Near Infrared Reflectance (NIR) spectroscopy has emerged as a promising alternative. In this study, a total of 200 maize seeds (100 seeds of sterile and 100 seeds of fertile line of KA 96-314) were analyzed using near-infrared (NIR) spectroscopy in the 900–1700 nm wavelength range with a DLP NIRNano device to evaluate the potential of classification of fertile and sterile maize lines by NIR spectroscopy. Three spectral data formats—intensity, reflectance, and absorbance—were evaluated under two preprocessing conditions: raw and a combination of first derivative (FD) and standard normal variate (SNV). For classification, two machine learning algorithms, support vector machines (SVM-C) and extreme gradient boosting (XGBoost), were applied. The results showed that preprocessing significantly affected model performance, with FD+SNV generally outperforming raw spectra. Also spectral data type has an effect on the model performance and reflectance data gave more successful results than the other data type. Among the tested models, SVM-C combined with reflectance data and FD+SNV preprocessing yielded the highest classification accuracy (Train set= 0.969, Test set=0.750), suggesting that NIR spectroscopy coupled with chemometric techniques can serve as a rapid, non-destructive method for CMS fertile/sterile seeds separation in maize breeding programs. These findings highlight the potential of NIR-based methods for streamlining CMS line development or genetic purity check in maize breeding studies.

Keywords: seed production; machine learning; classification; *Zea mays*

Genetic analysis of pollen morphology of inducer lines using the additive-dominance model of generation mean analysis

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Haploid induction efficiency is a key component of doubled haploid breeding systems, where pollen morphology serves as a useful phenotypic marker for selecting effective inducer lines. In this study, Generation Mean Analysis (GMA) was used to investigate the genetic control of pollen morphometric traits in two maize inducer lines, ADAIL-1 and Stock-6, along with their F₁ and F₂ generations. Morphological parameters including pollen area, perimeter, major axis, and minor axis lengths were measured using high-resolution image analysis. Additive-dominant genetic models were fitted based on the mean trait values of P₁, P₂, F₁, and F₂ generations. Mean trait values across generations indicated that F₁ and F₂ generally showed reduced trait expressions compared to the parental lines. For instance, mean pollen area decreased from 9468.5 µm² (P₁) and 9527.7 µm² (P₂) to 9337.1 µm² (F₁) and 8512.4 µm² (F₂). A similar trend was seen for perimeter and minor axis traits. Results showed that dominant effects were predominant in Area ($h = 167.52$) and Perimeter ($h = 16.07$), while additive effects were minimal or negative. Narrow-sense heritability (h^2) ranged from 0.208 (Perimeter) to 0.323 (Major axis), suggesting moderate heritability across traits. The highest genetic advance (GA) was recorded for Area (GA = 1145.94), followed by Perimeter (21.76) and Minor axis (8.99), indicating potential for phenotypic selection. Heterosis values revealed positive mid-parent and better-parent heterosis (MPH and BPH) for Perimeter (MPH = 3.18%, BPH = 2.17%) and Major axis (MPH = 1.00%), while Area and Minor axis showed slight negative heterosis, indicating trait-specific hybrid responses in F₁. These findings offer valuable insights into the inheritance of pollen traits and support morphological selection in maize haploid inducer breeding.

Keywords: pollen morphology; maize; haploid induction; additive-dominant model; heritability

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Chemical mutagenesis as a tool for improving elite inbred lines in maize

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Developing homozygous inbred lines is a fundamental step in any hybrid maize breeding program. However, the production and evaluation of inbred lines require extensive time, labor and multi-environment testing over several seasons. Given the low frequency of inbreds with strong combining ability, effective breeding programs must continuously generate and assess a large number of inbred lines, making the process both time-consuming and costly. While conventional improvement of elite inbred lines through backcrossing is possible, it often results in reduced combining ability. Alternatively, chemical mutagenesis provides a way to induce genetic variation within homozygous material, offering novel variation for further selection. In this study, B73 and Mo17 were used as model materials to evaluate the efficiency of chemical mutagenesis in improving key traits such as reduced vegetative period (from FAO-600) increased per se grain yield, and enhanced specific combining ability. After four growing seasons, all initial objectives were successfully achieved in the B73 line. In recent years, improving tolerance to extreme drought and heat has become a major breeding objective. Current efforts within the mutation-breeding program are focused on enhancing stress tolerance in Mo17 and other elite inbred lines.

Keywords: *mutagenesis; inbred lines; hybrids; improvement*

Overcoming the current challenges of haploid induction and increasing the efficiency of chromosome doubling

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During the last three decades, the doubled haploid (DH) technology has become the main approach for the production of maize homozygous lines within almost all world hybrid breeding programs. Climate changes (annual drought and heat problems), provoked us to start a project on the improvement of haploid inducers ten years ago. For the last two years, we have been testing our new inducers in comparison with the previous generation of inducers developed in 2010. We have managed to maintain the most important inducers' characteristics – the high rates of haploid induction (15-20%) and all three marker-gene systems allowing haploids to be selected at different stages. The biggest progress was achieved in the improvement of drought and heat tolerance - our main breeding goal. Currently, in the conditions of drought and extreme heat, we are able to work with new inducers – performing crosses with donors and maintaining their seeds, whereas the previous inducers have huge problems with their pollen production, silking and even to survive. A number of different chromosome-doubling methods have been elaborated lately. For the large production of DH lines, the most efficient approaches adapted to the certain conditions have to be used. This season, at our DH field, the rate of survival is over 80% and the male fertility is about 40%. Within our DH research project, we are testing new chemicals (alternatives of colchicine) for the chromosome - doubling treatments and applying different schemes of planting to increase the number of DH plants obtained per square meter.

Keywords: *DH project; inducers; haploids; chromosome doubling*

Genetic similarity analysis of maize doubled haploid lines by SSR markers

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Doubled haploid (DH) technology accelerates the development of homozygous lines and improves selection efficiency in maize breeding. In this study, seven DH lines developed from landraces and two reference inbred lines (B73 and Mo17) were evaluated to assess their genetic diversity and relatedness using 30 simple sequence repeat (SSR) markers distributed across the maize genome. DNA quantity and quality were measured, and high-quality genomic DNA was used for PCR amplification with primers selected for their polymorphism informativeness. Amplified fragments were scored as binary data, and key marker statistics—including allele number, polymorphism information content (PIC), and genetic diversity indices—were calculated. DNA concentrations ranged from 168.6 ng/μL to 237.9 ng/μL, with A260/A280 and A260/A230 ratios between 1.68–1.75 and 1.90–2.13, respectively. A total of 64 alleles were detected, with 1 to 6 alleles per locus. The mean PIC value was 0.30, indicating that the SSR markers provided moderate but sufficient polymorphism for genetic assessment. Additional parameters such as expected heterozygosity and Nei's gene diversity further supported moderate genetic variability. Cluster analysis based on Jaccard similarity coefficients and the UPGMA algorithm revealed distinct genetic groupings among the genotypes. The resulting dendrogram classified the DH lines into two main clusters. Genotypes such as BAF-DH10, BAF-DH15, BAF-DH8, and BAF-DH9 clustered together and were clearly separated from BAF-DH1, BAF-DH2, BAF-DH6, and the reference lines, suggesting the presence of divergent genetic backgrounds. This divergence highlights their potential as complementary parental lines in hybrid breeding. Overall, the findings offer valuable insights into the molecular characterization of DH maize lines and support their strategic integration into future breeding programs.

Keywords: *microstallite; polymorphism; genetic diversity; Zea mays*

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The logistic model in the analysis of dry matter accumulation in maize kernels

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In order to better understand the process of maize kernel dry matter accumulation under specific agroecological conditions, a two-year study (2022–2023) was conducted involving 10 maize hybrids of different maturity groups. Special emphasis was placed on modeling the dynamics of dry matter accumulation using the three-parameter logistic model (3P). The trial was organised as an RCB design, with four replications, at the Rimski Šančevi experimental field of the Institute of Field and Vegetable Crops in Novi Sad (IFVCNS). The dynamics of kernel dry matter accumulation was monitored every three days from silking to physiological maturity, and the time scale was expressed in growing degree days (GDD). Using the logistic model, maximum dry matter content (ASYM) and the point at which the kernel growth rate reaches its maximum (IP) were determined. Results revealed considerable differences among hybrids and between years. The early-maturing hybrid NS3023 showed the highest ASYM in 2022 (0.294 g), while in 2023, the highest ASYM was recorded in the late-maturing hybrid NS6061 (0.339 g). In contrast, hybrid NS5051 showed the lowest ASYM values in both years – 0.215 g in 2022 and 0.274 g in 2023. The IP ranged from 914.319 GDD (NS3022) to 1200.082 GDD (NS640) in 2022, and from 982.229 GDD (NS4006) to 1220.978 GDD (NS6061) in 2023. The results highlight the potential of the logistic model for analyzing kernel development and improving maize hybrid selection.

Keywords: *non-linear models; Growing Degree Days; Zea mays L.*

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Genomic mapping and prediction of silage traits in maize

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Improving silage quality and yield in maize hybrids is crucial for forage-based livestock systems. This study explores the genetic basis of 13 agronomic and silage-related traits—including plant height, forage yield, and nutritional quality—using a diverse panel of 225 maize hybrids evaluated under field conditions in Türkiye. Genotyping was performed using the DArTseq platform, yielding 4,525 high-quality SNP markers. Genome-wide association studies (GWAS) identified 51 significant loci, with key associations found for traits such as ear height, dry forage yield, and crude protein. Several loci were located near known candidate genes, offering insight into trait-specific genomic regions. To capture both additive and non-additive genetic effects, five multi-kernel genomic prediction models were evaluated, incorporating additive, dominance, and epistatic variance components. Additive effects explained a major portion of phenotypic variance for agronomic traits (up to 54%), while quality traits like protein and starch showed notable contributions from dominance and interaction effects. Genomic prediction accuracies were highest for traits like plant and ear height ($r \approx 0.80$), but lower and more variable for nutritional traits. Principal component analysis confirmed broad genetic diversity among parental inbreds and their hybrids, with hybrids displaying heterozygosity levels between 32–40%. Overall, the integration of GWAS and multi-kernel prediction provided novel insights into the genetic architecture of silage traits in hybrids and highlighted the importance of modeling non-additive effects for optimizing genomic-assisted selection. These findings advance the development of high-performing maize hybrids tailored for silage production.

Keywords: maize; silage; genomic; GWAS; prediction

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Cold stress tolerance during germination can help early sowing for mitigating climate change impact on maize yield

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The rate of global warming has dramatically accelerated in recent decades and the upward trend in Serbia is even higher than the global average – 0.60°C/decade for 1981-2017 period. Maize was recognized as the most endangered crop with projected net warming-related losses from 12% to 30% by the end of the century. Early sowing is one of adaptation strategies which could mitigate yield losses. Benefits of shifting sowing date forward are avoiding heat peak stress during flowering and grain filling, and improved water use efficiency through utilizing early-season moisture more effectively before summer droughts. However, this strategy bears risk of cold stress during germination which can damage seedlings and reduce stand establishment. Consequently, understanding mechanisms of cold tolerance and developing tolerant genotypes are required for realizing the full potential of early sowing. In LMGF (Laboratory for molecular genetics and physiology), research on physiological, biochemical and molecular response of maize seedlings to cold stress is in progress. Simple germination, phenotypic and physiological assessments were performed for streamlining genotype selection for more profound studies. Photosynthetic efficiency, membrane integrity, osmolyte accumulation and the activities of key enzymatic scavengers were assessed. Whole transcriptome profiling identified expression patterns of mRNAs and non-coding lncRNAs, miRNAs and circRNAs, as well as interactions between them. Further research is planned in two directions. First, adapted high quality protein maize (QPM) lines will be studied with the focus on photosynthesis, transpiration, hormonal regulation, ROS metabolism and antioxidant defense. Also, profiles of essential amino acids and other nutritive compounds will be established. Second, transcriptional biomarker panel, with integrated knowledge from transcriptomic, physiological and biochemical data, will be developed as a tool for rapid/reliable screening and more efficient maize breeding for cold tolerance. The expected outcomes include comprehensive insights into the plant's cold adaptation strategies, as well as biomarker application in advanced breeding.

Keywords: *early sowing; cold stress; germination; maize*

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Integrated GWAS and metabolomic analyses provide novel details about the genetic basis of free asparagine accumulation in durum wheat grains

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Free asparagine content is a key factor in acrylamide formation in wheat derivatives after high temperature processing. Therefore, the control of free asparagine levels is of interest in crop and food sciences. Our study aims to explore free asparagine natural variation in durum wheat grain to identify candidate genes controlling this trait. Two hundred and one durum wheat genotypes were selected from an international germplasm collection and sown in an experimental field for three years. Free asparagine content was measured on whole grain using an enzymatic assay. A genome-wide association study identified six associated quantitative trait nucleotides. Moreover, the whole grain metabolome of one-year samples was investigated to identify metabolic pathways associated to free asparagine accumulation. Specific enriched pathways involved in amino acids metabolism were detected and four candidate genes were identified. This study paved the way to characterize the genetic determinants of free asparagine accumulation in wheat grain.

Keywords: *free asparagine; durum wheat; GWAS; metabolomics*

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Comparative study of four genes' differential expression in maize leaves and roots during the v3 growth stage under low temperature stress

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Climate change triggers a series of adverse events that negatively impact maize production. Unexpected cold spells during early growth stages are one of them. Cold spells negatively impact plant metabolism, provoking decline in photosynthesis, delayed growth, and insufficient accumulation of crucial metabolites, which consequently reduce yield. Therefore, studying changes in maize plants induced by this factor could provide significant insights into the mechanisms underlying tolerance to low temperature stress. This study is focused on analyzing maize transcriptome response to low temperature stress at V3 growth stage in tolerant (Lt) and susceptible (Ls) maize inbreds. Plants were grown under low temperature (10°C/8°C) and control conditions (25°C/22°C) and sampled (leaf and root) 6h and 24h following the initiation of stress. After sampling, total RNA was extracted, cDNA library were constructed and NGS was done for the whole transcriptome followed by bioinformatics analyses. A notable amount of genes was found to be significantly differentially expressed in both analyzed sample types. Herein, findings of four genes included in different processes in maize plant that are significantly differentially expressed in both root and leaf samples is presented. These genes include: representative of the Myb-like transcription factor family (RVE1 homologue), crucial element of the Mitogen-Activated Protein Kinase (MAPK) cascade (NPK homologue), member of the Cytochrome P450s (CYPs) the largest family of enzymes involved in plant metabolism (CYP85A1 homologue), and a member of the serine carboxypeptidase-like proteins (SCPL50 homologue). Selected genes showed different expression profiles depending on maize inbred, treatment time point or type of sample (leaf or root). Specific expression patterns were found that do not consistently correspond to the expectations outlined in the literature concerning low temperature stress responses. Further studies and validation of gene expression are required to confirm their roles and elucidate the mechanisms of tolerance to the studied abiotic stress factor.

Keywords: maize; cold spells; transcriptomics; low temperature tolerance

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Marker assisted breeding for the maize nutritional enhancement

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Combined with traditional selection techniques, marker assisted breeding aimed at the maize nutritional enhancement has been established at the Maize Research Institute Zemun Polje. Over the years, commercial maize inbred lines of standard quality have been converted to quality protein maize (QPM), as well as to β -carotene rich (BCR) maize, adapted to temperate regions. These newly developed lines were used for developing high-yielding maize hybrids with improved nutritional benefit adapted to temperate regions. Simple sequence repeats (SSR) were efficiently used for direct selection of target gene (foreground selection) and for fast recovery of recurrent parent's genome (background selection). As gene-specific markers in foreground selection, phi057 and umc1066 were used for QPM lines, while crtRB1-3'TE marker was used for BCR lines. Background selection was performed with SSR markers evenly distributed throughout the genome. The recovery of the recipient genome in our best lines varied from 93% to 99% for QPM and from 90% to 99% for BCR. Biochemical evaluation of resulting improved hybrids showed increase of relevant nutritional parameters. Tryptophan content and tryptophan to protein ratio, an indicator of improved nutritional protein quality, were significantly higher ($p < 0.05$) in QPM hybrids compared to standard maize. Also, tryptophan was above 0.075%, which corresponds to the QPM threshold value. Similarly, β -carotene content in BCR hybrids was significantly higher ($p < 0.01$) in comparison with the standard hybrids (increase up to 78.11%). Furthermore, starch and lipid contents were significantly higher ($p < 0.01$) in improved hybrids. These results confirmed the success of the marker assisted conversion process, resulting in hybrids with improved nutritional value for use in feed industry.

Keywords: *maize; marker assisted breeding; nutritional enhancement; protein quality; β -carotene*

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Germination of adapted QPM inbred lines under different temperature patterns of cold stress

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Maize is one of the most important crops in the world, widely used as food and feed. Combined drought and high-temperature stress during flowering and grain-filling stages significantly reduce both grain yield and quality. Early sowing contributes to mitigation these negative climate change effects by avoiding unfavourable environment conditions during flowering. However, it implies germination and seedling growth at low temperatures. The aim of this study was to choose the temperatures that will be used to investigate impact of cold stress on germination of adapted maize inbred lines with high nutritive value (QPM). The data provided by the Republic Hydrometeorological Service of Serbia was analysed for the period from March 20th until April 10th, 2012-2023, on the territory of Belgrade. Four patterns of temperature fluctuation and duration were observed and used to design four congruent experiments. Maize seedlings were grown in a growth chamber (light intensity $700\mu\text{mol m}^{-2}\text{s}^{-1}$, humidity approx. 60%, photoperiod 12/12 h) for 14 days under control ($19^{\circ}\text{C}/11^{\circ}\text{C}$) and treatment (selected temperature sets) conditions. Germination parameters - germination percentage, germination index, germination energy, mean germination time and germination rate index, as well as root and shoot traits (length, fresh and dry weight), and vigour indices were determined. The temperature set selected for further investigations, which most comprehensively represents environmental conditions that enable successful germination, was as follows: $19^{\circ}\text{C}/11^{\circ}\text{C}$ for the first two days, then $20^{\circ}\text{C}/13.7^{\circ}\text{C}$, $9.4^{\circ}\text{C}/6.3^{\circ}\text{C}$, $6.5^{\circ}\text{C}/3.5^{\circ}\text{C}$ and $19^{\circ}\text{C}/11^{\circ}\text{C}$ for every three days, respectively. Further studies under controlled and field conditions will help better understand the effect of cold stress on maize seedlings, leading to the identification of promising lines for breeding high nutritive maize suitable for early sowing.

Keywords: cold stress; germination; temperature pattern; QPM

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Adjustment of maize production technology to variable climate parameters

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In regard to soil preservation as an important resource and maize cultivation under variable climatic conditions, reduced soil tillage is widely adopted. The experiment was settled down in 1978, and two periods, 2005-2014 and 2015-2024, were compared regarding the impact of soil tillage and fertilization regime on maize productivity. Maize hybrid (FAO 600) was grown with different tillage: no-tillage (NT), reduced (RT), and conventional tillage (CT), as well as N fertilization rate – 0 (F1), 180 (F2) and 240 kg N ha⁻¹ (F3), including rainfall conditions vs. irrigation, as the 3rd factor. Maize grain yield (MY) was measured after harvest and calculated at 14% of moisture. Data were processed by ANOVA, and Weibull analysis was conducted to evaluate the impact of meteorological factors on yield potential. MY was 7.8% and 16.0% higher in RT and CT than in NT during the 2005-2014 period, while the differences were 11.6% and 23.0% in the second period. Similarly, nitrogen application in F2 and F3 caused significant increase in MY by 26.0% and 33.3% in the first period and 23.1% and 27.4% in the 2015-2024 period. In average, MY was higher in the first (9.7 t ha⁻¹) than in the second ten-year's period (8.4 t ha⁻¹) for 34.7%. Irrigation increased MY in average from 8.3 to 11.1 t ha⁻¹ in the first period and from 7.2 to 9.7 t ha⁻¹ in the second period. Weibull analysis showed that NT has extremely high yield potential but only when all environmental conditions are extremely favorable (0.1 reliability). In less favorable growing conditions, yield decreases gradually, with increased differences between rainfall conditions and irrigation, especially when it comes to the RT and CT (reliability 0.9). This indicates that in agroecological conditions of Zemun Polje and slightly chalcaceous chernozem, CT is more convenient regarding maize yield.

Keywords: *reduced tillage; nitrogen; irrigation; maize; grain yield*

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Chemical composition of grain after mineral nitrogen fertilization of maize

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Maize is an important forage, food and technical crop. Regarding climate change, the efforts of breeders are aimed at creating hybrids with high adaptive and productive potential. Pesticide Application, Drift Reduction, Corn, Adjuvants, Collaboration. Appropriate agrotechnological practices are also important for successful maize cultivation. In intensive cultivation, a lot of mineral fertilizers are used, which are one of the most effective means to increase productivity. Mineral nitrogen fertilization is a prerequisite for higher maize yields. Poorly balanced fertilization is often the cause of severe yield reductions. The present work aimed to study the effect of two types of mineral nitrogen fertilizer (ammonium nitrate and urea) on the chemical composition of grain of maize, Kneja 561 hybrid. The standard growing technology was applied under dry farming conditions. Doses of 60 and 120 kg/ha of nitrogen (active substance) applied as ammonium nitrate and urea at the 6-8 leaf stage of maize were tested, including unfertilized control. Protein, starch, ash, and dry matter were determined using NIR spectroscopy for analyses. The results showed the chemical composition was more balanced under mineral nitrogen fertilization, with no statistically proven difference for the two mineral nitrogen fertilizers tested.

Keywords: *maize; nitrogen fertilization; chemical composition*

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Eco-Friendly ZnO nanoparticles for maize seed priming: Assessing seed germination and initial seedling growth

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Zn plays a key role as an essential micronutrient in the growth and development of maize plants. In this context, the application of nanotechnology has recently garnered significant attention in modern agriculture. Conventional chemical synthesis of Zn-based NPs typically involves toxic substances for particle reduction and stabilization. To address this, green synthesis methods have been developed using renewable and eco-friendly raw materials such as plant extracts, starch, microorganisms, and yeast, as alternatives to hazardous chemicals. ZnO NPs were synthesized via green hydrothermal synthesis in the presence of starch, which serves as a reducing and stabilizing agent. This method allowed precise control of particle size, limited growth, and prevented aggregation. The resulting NPs were applied as maize seed priming agents at four concentrations (40, 80, 100, and 160 ppm) to three different hybrids NS 6005, NS 11410, and NS 12402, to assess their impact on maize seed quality and initial growth. Hydro- and nanopriming treatments were performed at room temperature for 16 h followed by thorough rinsing and air-drying for 48 h. The experiment was conducted under optimal laboratory conditions in accordance with ISTA (2025) standards. Among the tested concentrations, 80 ppm ZnO NP resulted in the most favorable physiological response, especially in hybrid NS 6005. Although ZnO NP did not affect seed germination because the seed lot was highly vigorous (99%), they significantly improved the shoot and root lengths of maize seedlings compared to the control by 28.35% and 19.69%, respectively. In addition, 80 ppm ZnO NPs enhanced shoot and root biomass accumulation, with fresh mass increases of 17.62% and 37.06%, and dry mass increases of 15.67% and 49.87%, respectively. These findings suggest ZnO NPs synthesised via this green method can be effectively used as a priming agent, highlighting their potential in sustainable agriculture.

Keywords: maize; nanopriming; seed quality; Zn-based nanoparticles; initial plant growth and development

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The effect of the application of biostimulators on the seeds of ZP lines and hybrids of maize

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Biostimulators are frequently used in modern agricultural production to promote crop development, improve stress tolerance (drought, disease and temperature fluctuations), better utilization of nutrients from the soil and increase yields. They can be applied to seeds, incorporated into the soil or applied foliarly. In this experiment, the effect of 11 biostimulators applied to the seeds of ZP maize genotypes (5 inbred lines and 5 hybrids) was tested. Germination on two different substrates: between filter paper (BP) and soil in rolls (S), and comparison with the control was monitored. Analysis of variance showed that there was no significant difference in the percentage of germination between the two substrates (BP 95%, and S 93%). A significant difference in the percentage of germination was observed between the control in the S (97%) and the seeds treated with biostimulators, on the same substrate (93%). Germination values of seeds treated with biostimulators and controls on filter paper were identical (95%). On average, the germination of maize hybrids (96%), as expected, was significantly better than the average germination of maize inbred lines (91%). Tested genotypes responded the best to the biostimulator No 7, with an average germination of 96%, and significantly differed from the seeds treated with biostimulator No 9 (89%). The seeds of hybrid 1 had the most robust root system during the evaluation of germination, when treated with biostimulator No 4 in soil, and the highest percentage of germination (100%) in the same substrate when treated with biostimulator No 7. The experiment showed that the germination test of seeds treated with biostimulators can be done on both substrates. In general there was no significant increase in the germination of tested seeds treated with biostimulators, but there were some combinations of biostimulators and genotypes with positive reactions manifested by robust root system and germination percentage.

Keywords: *maize; seeds; biostimulators*

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Grain yield after monoculture and intercropping of maize

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Climatic change has created challenges for the agricultural sector, adding to pressures on global agricultural and food systems. The increase in the frequency and duration of droughts requires a strong adaptation of agricultural crops, as well as suitable technologies for their cultivation. Considerable attention can be given to croping strategies such as intercropping systems. Intercropping is an important cropping system that has benefits in improving crop yields during climate change. The present work aimed at to study the yield of grain after monoculture and intercropping of maize. Field experiment was carried out at the Maize Research Institute - Knezha, Bulgaria. Maize hybrid Knezha 561 (Bulgarian selection) was grown as in monoculture. The standard technology of cultivation was applied under dry farming conditions. The mixture of pea:oat in 70:30 ratio was used for the intercropping. Grain yield of maize (14% moisture) was recorded. Results showed that mineral nitrogen fertilization effected on the grain yield of maize, with higher values being recorded at a dose of 12 kg/da for both nitrogen fertilizers. Higher grain yield was obtained with intercropping of maize as compared to the monoculture. Relatively lower variation was found when using ammonium nitrate compared to urea.

Keywords: *maize; intercropping; fertilization*

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Conducting Horizon EU Project GOOD - The agroecologically based weed management in maize

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Within the “Agroecology for weeds-GOOD” Project, Living Lab focusing on maize weed control was established in Serbia. The reduction of herbicide use and risk has become major policy targets of EU, aiming to promote the transition to sustainable and resilient farming systems. The mission of the GOOD project is to evaluate the effects of the multidisciplinary approach - Agroecological Weed Management (AWM) system and support farmer’s acceptance of this concept in annual and perennial crops in different agroclimatic conditions in Europe. Maize was grown within Living Labs in organic and conventional system. In the first experimental year, three cover crops (CC): oat, rye, and winter vetch were sown in autumn, and incorporated into the soil after the termination in spring. The control was without CC. They were followed by maize as the main crop in which various weed management practices were evaluated: mechanical weeding, mulching, false seedbed, bio-based herbicide, and untreated control for the organic system; as well as in the conventional system: herbicide application in either recommended or reduced rate (0.5x rate), mechanical control, bio-based herbicide, and the control plot (untreated). The efficacy of combined application of CC and weed control measures was assessed on a weekly basis, up to four weeks. The higher number of weed species was present in plots with rye and vetch, in organic maize, thus supporting weed biodiversity, while similar results in conventional maize were present only in combination of vetch + weedy check. Nevertheless, rye and oat significantly reduced weed density and dry mass compared to no cover crop plots, particularly in combination with mechanical weed control in organic maize. In conventional maize, all three cover crops were highly effective in reduction of weeds, especially in combination with herbicides. The results indicate that is challenging to support farmers to adopt agroecological strategy for weed control.

Keywords: agroecology; EU project; weed control; maize

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Valorization legumes related agrosystem services: Living Lab in Serbia

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VALERECO aims to accelerate the adoption and recognition of legume crops as key drivers in the shift toward sustainable, productive, climate-resilient, and environmentally friendly farming systems. By assessing both the environmental and economic contributions of ecosystem services (ES) provided by eight major and four minor or underutilized legume species, the project supports greater crop diversification across the EU and associated countries. To put this into practice, VALERECO established nine Living Labs across Europe to (1) design behavioural strategies that promote legume uptake in both production and consumption, (2) conduct participatory field trials to evaluate legume performance in diversified systems, and (3) co-develop technical solutions that are both economically and environmentally viable. One of the Living Labs is located in Serbia, and the main legume crops are soybean and chickpea. Furthermore, the trial included field peas as a winter cover crop and intercropping barley (pure and strip). Field monitoring of winter cover crops, particularly field pea, was conducted to evaluate their establishment and performance under local agro-ecological conditions as well as selected mechanical weeding measures. Field trial preparations, including soil sampling, sowing, and monitoring, were successfully executed. The trial will last for three years, following 3D diversification scheme. Within the Living Lab, a stakeholder survey was conducted (consumers, farmers, and retailers). In the following period, the trainings and demo events will be organized. The project's results are and will be widely disseminated through the creation of a Digital Legume Information Hub (DLIH), a Decision Support System (DSS) for farmers and advisors, and an open-access E-learning Platform for capacity building.

Keywords: *legumes; ecosystem services; living labs*

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Sustainable agricultural strategies for optimizing soybean and millet forage biomass productivity

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In response to increasing livestock populations and the need for more ecological agricultural practices, environmentally unsafe methods are being replaced by approaches that enhance forage security. Intercropping and biofertilization are recognized as sustainable strategies to address prevention of yield loss and malnutrition. This study evaluated the effectiveness of soybean-common millet intercropping and the biofertilizer Coveron (BF) on forage biomass productivity, using yield, land equivalent ratio (LER) and concentrations of P, B, Fe and Zn – key nutrients often limited in calcareous soils. Three intercropping patterns were tested: AR - alternating rows, AS1 - alternating two-row strips, AS2 - alternating two-row soybean with four-row millet strips, alongside sole crop controls. Results revealed that AS1 achieved LER > 1 for both fresh and dry biomass, despite reduced millet yield. BF had no significant effect on forage biomass and LER values. Sole soybean had the highest P concentration in both vegetative and reproductive part, while AS1 increased B and Zn accumulation in vegetative part of biomass with minimal P reduction and insignificant change in regard to Fe concentration. In millet, AR combination significantly affected P accumulation, increasing its level in reproductive part, simultaneously with reduced B concentration by all intercrop combinations. AS1 improved Fe level in both parts of millet biomass, while Zn showed different accumulation and translocation, depending on planting pattern. BF positively influenced P accumulation in soybean pods and B in millet panicle, but negatively affected P and Zn accumulation in millet vegetative part. Accordingly, intercropping soybean and common millet in alternating two-row strips represents a feasible sustainable solution for enhancing land use efficiency along with the accumulation of key nutrients in forage biomass.

Keywords: *intercropping; biofertilizing; yield; land equivalent ratio (LER); elements*

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Weed management and yield performance in edamame soybean

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Edamame soybean is a nutritionally rich vegetable crop widely recognized as a healthy food in the human diet. Edamame is harvested at the BBCH 76, when pods and beans are still green. Weed control strategies essential for achieving optimal edamame yields. This study aimed to evaluate edamame yield performance and weed control under the influence of herbicide treatments: H0 – untreated control; H1 – pre-emergence application of metribuzin (350 g ha⁻¹) and s-metolachlor (1334 g ha⁻¹); and H2 – H1 treatment followed by post-emergence application of imazamox (44 g ha⁻¹) and cycloxydim (200 g ha⁻¹). Field trials were conducted with two edamame varieties: Chiba Green (V1) and Midori Giant (V2) during 2022 to 2024 growing season. Herbicide treatments induced significant differences in weed fresh and dry biomass. Regarding the year×herbicide interaction, weed biomass under H0 was significantly reduced in 2024 (1063.0 g m⁻²), while it remained stable in 2022 and 2023 (average 2569.8 g m⁻²). In 2022 the dominant weed species were *Chenopodium hybridum* and *Solanum nigrum*, whereas in 2023 and 2024 *Chenopodium album* and *Sorghum halepense* prevailed. H1 was the most effective in 2023, (with no significant difference from H2), but failed in 2024, with biomass levels exceeding those in the control (1276.7 g m⁻²). H2 consistently provided effective weed suppression across all seasons. The herbicide×variety×year interaction significantly affected pod yield. Midori Giant outperformed Chiba Green by an average of 4.5 t ha⁻¹. The highest yields were recorded in 2022 (10.2 t ha⁻¹), followed by 2023 (8.2 t ha⁻¹) and 2024 (6.6 t ha⁻¹). Combined application of pre- and post-emergence herbicides (H2) ensured consistent weed control and yield stability, highlighting its importance for reliable edamame production. Application of only pre-emergence herbicides may be sufficient, but only under favorable meteorological conditions.

Keywords: Edamame soybean; weed control; weed biomass; pod yield

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Mineral composition of the common winter wheat grain grown in different tillage and nitrogen levels

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The content of mineral elements is a fundamental component of wheat grain quality and varies depending on applied cultural practices. The aim of the study is to examine the impact of wheat production methods on the composition of mineral elements in the grain. The field experiment was conducted in completely randomized blocks on luvisc chernozem soil of the Faculty of Agriculture "Radmilovac". The variety of common soft wheat Ilina (*Triticum aestivum* ssp. *vulgare*), was cultivated in three tillage systems - conventional, mulch and no-tillage. In addition to basic cultivation fertilization treatment with NPK fertilizers in the autumn, two different amounts of N in top dressing (60 and 120 kg/ha N), were added in spring. Standard measures in wheat production were applied and the harvest was carried out at the end of June. Concentrations of 18 elements (As, Al, Ba, Ca, Co, Cr, Cu, Fe, Hg, K, Mg, Mn, Ni, S, Sr, P, V and Zn) in wheat grain samples were determined by means of inductively coupled plasma with optical emission spectrometry (ICP-OES). The results indicated that the concentrations of the studied elements in the wheat grain were significantly affected by the tillage systems and fertilization rates ($p < 0.001$), as well as the interaction of these two factors. The lowest content of the examined macroelements (K=3052mg/kg and P=3617 mg/kg), as well as the highest concentration of microelements, such as (Cu=5.104 mg/kg and Ni=0.517 mg/kg), was observed in the grain produced in the conventional tillage. The highest concentrations of Fe 48.28 mg/kg, Ba 2.991 mg/kg, and Mn 24.4 mg/kg were detected in the grain treated with 60 kg/ha N. Reduced tillage and reduced nitrogen fertilization had significantly higher positive effect on the concentration of nutritionally important minerals (Ca, Fe, K, Mg, P and Zn) in respect to the conventional tillage with higher levels of nitrogen. Results indicate that in semi-arid climate reduced soil tillage could serve as an important part of sustainable cropping system to facilitate food security.

Keywords: grain of wheat; macroelements; microelements; ICP-OES

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Sustainable precision nitrogen fertilization in durum wheat: Insights from NNI and NDVI approaches

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Durum wheat is a key staple crop globally, with increasing nitrogen (N) fertilizer use driven by the need to sustain rising food demand. However, inefficient N management not only reduces resource use efficiency but also contributes to environmental degradation. Precision agriculture technologies, particularly variable-rate fertilization (VRF) informed by satellite imagery, offer the potential to optimize N application while maintaining yield and quality. This study evaluated the effectiveness of different N fertilization strategies in durum wheat over four consecutive growing seasons (October 2018–July 2022) in Asciano, Siena, Italy. Four approaches were compared: a conventional uniform N rate, and three VRF strategies based on Sentinel-2 L2A spectral bands. The VRF approaches included one guided by the Nitrogen Nutrition Index (NNI), a proportional NDVI-based estimate (NDVIH), and a compensative NDVI-based estimate (NDVIL). Grain yield, quality parameters (including protein content and technological properties), N fertilizer use efficiency (NfUE), and economic performance were assessed. Results showed that the NNI-based approach achieved significant reductions in N input without negatively affecting grain yield or quality. This method improved protein partitioning and dough rheological properties, critical for pasta production and other end-uses. Moreover, the NNI consistently delivered the highest NfUE among all treatments. Economic analysis revealed that the NNI approach offered lower social costs and higher returns on investment compared to both the uniform rate and the other VRF methods. These findings demonstrate that satellite-guided VRF, particularly the NNI approach, can simultaneously enhance economic viability and environmental sustainability in durum wheat production. By optimizing N inputs to actual crop requirements, this technology reduces resource waste, mitigates environmental impacts, and supports long-term agricultural resilience. The study provides practical evidence supporting the adoption of NNI-based VRF as a sustainable fertilization strategy in durum wheat cultivation under Mediterranean conditions.

Keywords: *variable-rate fertilization; life cycle assessment; N fertilizer use efficiency; social cost; dough technical properties*

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Effects of foliar selenium application on grain yield and nutritional quality of common wheat

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Selenium (Se) is an essential micronutrient for human health, yet its deficiency is widespread worldwide due to low concentrations in staple crops such as common wheat (*Triticum aestivum* L.). Increasing Se content in wheat grains through foliar application is a promising strategy to address this deficiency without compromising yield. This study evaluated the effects of different Se rates and application timings on grain yield and quality in two bread wheat varieties (Bologna and Sieve) grown under rainfed conditions. Field trials were conducted over two growing seasons (2020–2022) at two locations in Italy, using a split–split plot design. Selenium was applied at five rates (0, 2.57, 5.14, 10.27, and 20.54 g ha⁻¹) and at two growth stages (flowering and watery ripe). Measured parameters included grain yield, protein content, starch concentration, free asparagine levels, and phytic acid (Phy) content. Data were analyzed using a mixed-effects linear model to assess the interactions between varieties, Se rates, and application timings. Results showed that Se application did not affect grain yield in either variety. However, significant effects on grain quality were observed, with responses varying by variety. Se application reduced free asparagine and phytic acid contents, with the decrease in free asparagine being more pronounced in Sieve. Additionally, the molar Phy:cation ratio decreased in both varieties, indicating improved mineral bioavailability. Nevertheless, high Se rates were associated with reduced protein and starch contents. In conclusion, foliar Se application proved effective in lowering anti-nutritional factors such as phytic acid and free asparagine and in improving Se bioavailability, while having no effect on grain yield. However, reductions in protein and starch contents at higher Se rates highlight the need to tailor application strategies to specific wheat varieties. Such optimization could enhance the nutritional quality of wheat-based foods and contribute to addressing Se deficiencies in human diets.

Keywords: agronomic practices; crop nutrition; Se foliar application; micronutrient enrichment; trace elements

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Effects of agroecological conditions and agrotechnical practices on sunflower grain characteristics

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The production of sunflowers has multiple economic importance in the world. The stability of sunflower production significantly depends on abiotic stress. The occurrence of stress is significantly influenced by global and regional climate change, which will be increasingly pronounced in the near and distant future. Consequently, it has been decided to study the impact of agroecological conditions on grain yield, oil content and oil yield of sunflower grains. The object of study was three sunflower hybrids (NS-H-111, Velja and Bačvanin), which had been developed at the Institute of Field and Vegetable Crops, Novi Sad. Small-plot field trials were conducted in the period from 2007 to 2011, in three locations (Bačka, Banat and Srem). The experiments were conducted in a randomized block design with four replications. Sunflower grain yield, oil content and oil yield were determined. The obtained results indicated that the grain yield of sunflower was not significantly affected by genotype. However, locality and year significantly affected the grain yield. Localities Banat and Srem showed significant difference. The oil content was significantly affected by the studied hybrids, location and year. Factor analysis confirmed that the oil yield of sunflower mostly depends on the oil content in seed. The studied hybrids, location and weather conditions significantly affected the sunflower oil yield.

Keywords: *sunflower; agroecological conditions; yield; grain*

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Maize – winter wheat rotation and pre-emergence herbicides: a successful case of Integrated Weed Management

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Crop rotation is a simple and effective part of an Integrated Weed Management system, which makes it possible to rotate herbicides with different modes of action. Besides all the known advantages and benefits, it is still not widely used in maize production in Serbia. The aim of this research was to test the benefits of growing maize in crop rotation with winter wheat compared with continuous cropping, combined with pre-emergence herbicide application. Field trials started in 2009, and five maize-winter wheat rotations have been completed. Weeds were controlled with a herbicide mixture of isoxaflutole and S-metolachlor, applied at either the full label rate or half rate, while one plot was kept weed free (manually), and one was a control. Integrating crop rotation and PRE herbicides decreased the biomass of weeds and their density by 98% and 99%, respectively. In continuous maize, perennial weeds became dominant (mostly johnsongrass) after the first two years, while not the case in maize-wheat rotation. Crop rotation significantly influenced maize productive parameters, decreasing the variation in leaf area index and grain yield, and increasing values with the number of cycles. The biggest differences in the analyzed parameters were observed in 2015, 2017, and 2019, indicating many positive long-term benefits of crop rotation on maize leaf area index and grain yield.

Keywords: corn; herbicides; IWM; continuous cropping

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***Fusarium* ear rot resistance in popcorn inbred lines**

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Fusarium ear rot disease in popcorn is caused by *Fusarium* spp. and one of the most prevalent species is *F. verticillioides*. Higher disease severity mostly results in higher toxin contamination. In order to identify sources of resistance to *Fusarium* ear rot in popcorn elite germplasm, two groups of popcorn inbred lines were evaluated in natural and artificial ear infection: a) 30 old inbred lines including the parents of all previously registered OS hybrids (L1- L30) and b) 25 newly developed lines obtained from F1 hybrids of unknown origin (from different seed companies) that showed the best performance for yield, popping volume and ear rot resistance in multi-year trials (L31-L55). Field trial was conducted at the experimental field of Agricultural Institute Osijek, Croatia in 2023 according to randomized complete block design in three replications. Each plot consisted of two rows 5 m long. Primary ears in second row were inoculated (6 days after 50% silking) with *F. verticillioides* by injecting conidia suspension (10^6 conidia/mL) through the husk into the kernel wounds. At harvest time, ears were rated for *Fusarium* ear rot severity (% of kernels with visible disease symptoms). In natural infection, disease severity was low and ranged from 0 to 19 % and from 0 to 10% in old and new inbreds, respectively. With kernel inoculations a general increase in disease severity occurred. Ear rot ranged from 2 to 53% in old inbreds and from 2 to 42% in new inbreds. A moderate positive correlation was obtained between natural and artificial infection ($r=0.52$). However, some inbreds showed high resistance in natural but low resistance in artificial infection indicating usefulness of artificial infection in breeding for ear rot resistance. New inbreds with highest resistance (L31, L32) are included in testcrosses for commercial hybrids and could be used as sources of resistance to *Fusarium* ear rot in further popcorn breeding.

Keywords: popcorn; inbred lines; *Fusarium* ear rot

Effects of global climate change on toxigenic *Fusarium* species infecting maize and small grains in Serbia

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Global climate changes have caused variability in agroclimatic conditions, which may promote the accumulation of higher concentrations of mycotoxins in cereal grains during the growing season and economic losses in production, as well as an increased risk to human and animal health. These reasons and the recent identification of new toxigenic *Fusarium* species in Serbia in the last few years indicate the need for constant monitoring of mycopopulations during grain production. Since the phenotypic differences were not clear, a polyphasic approach was used to differentiate these species. In this research, the GCPSR (Genealogical Concordant Phylogenetic Species Recognition) approach based on the phylogenetic agreement of three independent genes was applied. In this way, the separation of species within the *Fusarium graminearum* complex (Fg) was made possible, which was not possible based on morphological characteristics. Using the TEF-1 α gene, revealed that it was not informative enough to separate all the species present within the Fg complex, although differences were detected based on the mentioned gene, even in a few nucleotides between isolates of the same species. Based on the TEF-1 α gene, isolate 1339/2 was identified as *F. vorosii*, while all other isolates were identified as *F. graminearum* s.s. Additional genes, β -tubulin and histone H3, were included in further analyses. Genes TEF-1 α and β -tubulin enabled detection of *F. graminearum* s.s. and *F. vorosii* species, while *F. boothii* could not be separated from *F. graminearum* s.s. Based on the histone H3 gene, all three species (*F. graminearum* s.s., *F. vorosii* and *F. boothii*) were identified. Given the differences obtained using three different genomic regions in this study, multilocus DNA sequences of three genes (TEF-1 α , β -tubulin and histone H3) were used for further phylogenetic analyses and tree construction.

Keywords: maize; small grains; *F. graminearum*; *F. vorosii*; *F. boothii*

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Sugar and free amino acid response of wheat under *Fusarium* infection

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Fusarium head blight (FHB) is one of the most dangerous diseases that significantly impacts wheat production. Besides negative impact on grain yield, technological traits of grain can be negatively affected because grain composition is altered by the fungus indicated by the degradation of cell walls, starch granules and storage proteins. We investigated and analysed the changes of sugars and free amino acid content induced by FHB colonization in six wheat varieties with differing in *Fusarium* resistance. The concentration of sugars and free amino acids was determined using a high performance liquid chromatography device. The significant increase of the total content of free amino acids was observed in wheat under FHB infection due to their susceptibility. The most FHB susceptible Tika Taka significantly increased it by 46%, while the most resistant variety Vulkan significantly decreased it by 16%. Also, the most FHB susceptible variety Tika Taka showed significant increase of the average total amount of fructose, glucose, maltose, total sugars, and total reducing sugars in the flour from grains from FHB infected treatment, compared to non-treated flour. To conclude, FHB results in a reduction in grain yield, grade and end-use quality, especially in FHB susceptible varieties, as consequence of hydrolysis of sucrose into fructose and glucose, together with increase in free amino acids which impairs technological and rheological quality.

Keywords: *free amino acids; Fusarium; sugars; wheat*

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Cold-stressed soybean susceptibility to Charcoal Rot

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This study investigated the susceptibility of early-vegetation cold-stress soybean to Charcoal Rot (*Macrophomina phaseolina* (Tassi) Goid) previously isolated from industrial hemp (*Cannabis sativa* cv. Fibranova). Nine soybean cultivars (six of them creation of soybean breeding program at the Agricultural Institute Osijek, and three introduced) were subjected to three-day cold stress in a walk-in growth chamber starting when the soybean had a fully developed first trifoliate (V2 stage), i.e. 20 days after sowing (DAS). Tested cultivars belong from 00 to 0-I maturity group. The infection was performed on cold-stressed soybeans and soybeans grown in optimal conditions at 30 DAS by cut-stem inoculation technique. The infection lesion length was measured every three to four days for three weeks. Cold stress significantly affected the intensity of the symptoms compared to soybeans grown in optimal conditions for all except the S9 cultivar. There were also significant differences between cultivars in their infection response. Tested cultivars responded differently to artificial infection under optimal conditions, and differently under cold-stress conditions.

Keywords: abiotic stress; *Macrophomina phaseolina*; multi-host pathogen; genotype effect

Novel toxigenic species of the genus *Aspergillus* on maize and small grains in Serbia

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Species of the genus *Aspergillus* are significant fungal pathogens that can colonize cereals, and pose a threat to food safety due to their ability to produce mycotoxins. In recent years, three potentially mycotoxigenic species (*Aspergillus parasiticus*, *Aspergillus tubingensis* and *Aspergillus welwitschiae*) have been identified for the first time in Serbia on maize and small grains. Their emergence is linked to the increasingly evident effects of climate change, particularly rising average temperatures, more frequent droughts, and extreme weather conditions during the growing season. These climatic shifts create favorable conditions for the development of thermophilic fungi such as *Aspergillus* spp., not only under post-harvest storage conditions, as was previously reported, but also directly in the field. The identification of new species within the Flavi and Nigri sections highlights ongoing changes in the mycobiota of cereals, requiring further research and adaptation strategies. Such strategies should include the selection of tolerant hybrids and varieties and improved agronomic practices to mitigate the impact of fungal pathogens, maintain crop yields, and ensure food safety.

Keywords: Maize; small grains; *A. parasiticus*; *A. tubingensis*; *A. Welwitschiae*

Acknowledgment: This study was supported by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia (grant no. 451-03-136/2025-03/200040).

Comparative studies of the mycobiota of maize and wheat seeds in the Republic of Srpska

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In the Republic of Srpska (RS), maize is one of the leading cultivated crops, with 141,021 ha under production, playing an important role in livestock farming and the processing industry. Wheat, although grown on smaller areas (50,040 ha), also has economic significance. To improve knowledge of the mycobiota associated with these crops, seed samples of maize stored for 6 months and wheat stored for 9 months were analyzed. The samples were taken from three different regions of the RS (Banja Luka, Doboje and Bijeljina regions). Standard phytopathological methods were applied for seed incubation, fungal isolation, and identification. For each sample, 30 surface-sterilized seeds were incubated on potato dextrose agar. After 7 days under laboratory conditions, both macroscopic (colony properties) and microscopic characteristics (*in situ* and all morphological structures) of the fungi were examined. The results revealed more than 18 fungal species, most of which were found on both maize and wheat seeds, although with varying frequencies. These species included (maize/wheat): *Alternaria* spp. (2.0%/35.0%), *Aspergillus flavus* (9.8%/2.5%), *A. niger* (2.9%/7.1%), *Fusarium graminearum* (2.0%/10.6%), and *F. verticillioides* (62.6%/1.37%). Other fungi detected on both hosts were *Cladosporium* spp., *Epicoccum nigrum*, *Penicillium* spp., and *Sarocladium zeae*. Additional species such as *F. proliferatum*, *F. sporotrichioides*, *F. subglutinans*, *Nigrospora oryzae*, and *Penicillium oxalicum* were recorded on maize seeds. In contrast, *Arthrinium phaeospermum*, *Bipolaris sorokiniana*, *F. avenaceum*, and *F. poae* were identified on wheat seeds. These findings highlight that special attention should be directed to *F. verticillioides* and *A. flavus* in maize, and to *Alternaria* spp., *F. graminearum*, and *A. niger* in wheat production. These fungi are important due to their role in seed, seedling, stalk, and ear/spike diseases, which can negatively affect both yield and grain quality. An additional concern is their ability to biosynthesize mycotoxins, thereby posing risks of grain contamination and food safety issues.

Keywords: maize; wheat; grain; fungi; incidence; Republic of Srpska

Harnessing virus resistance in balkan durum wheat to advance sustainable production

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Durum wheat production is increasingly threatened by agronomic constraints and viral diseases such as soil-borne cereal mosaic virus (SBCMV) and soil-borne wheat mosaic virus (SBWMV), both transmitted by *Polymyxa graminis*. These persistent pathogens pose significant risks to yield stability, especially under changing climatic conditions. Breeding progress is constrained by the limited genetic diversity of modern germplasm and the scarcity of resistance sources. Evidence from barley and bread wheat suggests the Balkans may represent a secondary center of resistance to soil-borne viruses. The newly developed Balkan Durum Panel, composed of old landraces cultivated in the Western Balkans until the 1970s, exhibits distinctive traits such as tall plant stature, long awned spikes, and tolerance to diverse biotic and abiotic stresses (Pavićević, 1982), making it a valuable resource for breeding. Genotyping-by-sequencing (GBS) generated 4,409 high-quality polymorphic SNP markers after stringent quality filtering. Principal component analysis (PCA) revealed four genetically distinct groups, while identity-by-state (IBS) analysis identified closely related genotypes (IBS > 0.95) and duplicates (IBS = 1; RD = 0) as reported by Velimirović et al. (2023). ADMIXTURE analysis confirmed a structured population, with cross-validation indicating K = 10 as the optimal number of subgroups. These findings underscore the importance of selecting genetically distant accessions for subsequent genome-wide association studies (GWAS). Given the increasing vulnerability of pasta wheat production to combined biotic and abiotic stressors, the Balkan Durum Panel holds substantial promise for identifying novel alleles and genes. Beyond virus resistance, its genetic diversity offers a strategic pathway toward developing more resilient and sustainable durum wheat production systems.

Keywords: Balkan Durum Panel; genotyping-by-sequencing (GBS); soil-borne cereal mosaic virus (SBCMV); soil-borne wheat mosaic virus (SBWMV); *Polymyxa graminis*

Impact of pest control strategies on oxidative metabolism in maize hybrids

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Phytochemical compounds and their antioxidant activity play a crucial role in maize's response to biotic and abiotic stresses, influencing nutritional quality and contributing to phenotypic variation essential for yield improvement and breeding strategies. However, pesticide-induced cytotoxicity and phytotoxicity can elevate reactive oxygen species (ROS) levels, leading to lipid peroxidation and oxidative stress. This study aimed to evaluate the antioxidant capacity of maize hybrids by measuring carotenoids (lutein + zeaxanthin, and β -carotene), tocopherols (α -, γ -, and δ -), total phenolic content (TPC), and DPPH radical scavenging activity in maize grain following exposure to various pesticide treatments. Three maize hybrids (ZP 457, ZP 5601, and ZP 606), developed at the Maize Research Institute "Zemun Polje," were treated under field conditions with five foliar insecticides (FT1–FT5) and two seed treatments (ST1 – insecticide; ST2 – fungicide), in 2022. Among the tested hybrids, ZP 457 showed the most notable average increases across all treatments, with δ -tocopherol and γ -tocopherol levels rising by 125.3% and 43.8%, respectively. In contrast, TPC exhibited the most significant average decreases—38.2%, 37.4%, and 32%—across all hybrids and treatments. Additionally, divergent trends in the percentage changes of certain bioactive compounds were observed among the hybrids in response to pesticide applications. For instance, β -carotene content showed considerable average changes in ZP 457 and ZP 5601, with increases of 21.5% and 16.05%, and decreases of 13.5% and 16.02%, respectively. These results suggest that a single parameter cannot fully capture the impact of pest control strategies on oxidative metabolism in maize, and that hybrid-specific responses should be considered.

Keywords: *Zea mays* Z; total phenolic content; tocopherols; carotenoids; DPPH

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Changes in bioactive compounds of maize hybrids grains under different insecticide treatments

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Maize, one of the top three cereal crops impacted by abiotic stress, thus becomes more vulnerable to the occurrence and intensity of European corn borer (ECB) attacks, one of the most important maize pests. The present study investigated the response of six maize hybrids (ZP 427, ZP 434, ZP 555, ZP 600, ZP 606, ZP 666) to three insecticides—chlorantraniliprole (C), bifenthrin (B), and a combination of lufenurool, chlorpyrifos, and cypermethrin (LCC)—during the second generation flight in the experimental field of the Maize Institute Zemun Polje, Serbia, during 2019 and 2020. It aimed to examine the variation in bioactive compounds, such as phenolics, tannins, and flavonoids, in the grains of different maize hybrids after insecticide treatments, compared to untreated controls. These compounds provide a range of nutritional and functional benefits, particularly due to their antioxidant properties. The results revealed that phenolic content increased in all hybrids under the C treatment compared to the untreated control. However, tannins and flavonoids increased in four hybrids, except ZP 427, which displayed a decrease in tannin content with C, while ZP 666 experienced a reduction in flavonoid content. In contrast, in the B treatment, levels of tannins and phenolics decrease in all analysed hybrids, while flavonoids decreased across five hybrids, except in ZP 434. A similar decreasing trend was observed for these bioactive compounds under LCC treatment, where the content of tannins and flavonoids increased only in ZP 606, and the content of phenolics increased in both ZP 606 and ZP 555. Changes in those phytochemicals are connected to the plant's defence mechanisms and responses to stress, as well as its overall physiology. Findings from this study demonstrate that insecticide application affects the bioactive compounds of maize grain.

Keywords: *Ostrinia nubilalis* Hübn; phenolic; tanni; flavonoid compounds; maize

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Challenges in modern maize seed production

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Upcoming climate change, particularly high temperatures and the lack of precipitation during the growing season, have a negative impact on maize yields in both commercial and seed production. Additionally, the seed production sector is facing increasing difficulties in sourcing sufficient labor. Through its breeding program, the Maize Research Institute has developed inbred lines that ensure high and stable yields in seed production. In recent years, a growing number of consecutive seasons have been characterized as unfavorable for maize seed production, including the years 2021, 2022, and 2023. During this period, the average seed yield of the most widely produced hybrids ranged from 3,308 to 3,654 kg/ha for ZP 427, and from 1,497 to 3,104 kg/ha for ZP 606. The female components of these hybrids exhibited high and stable yields, while the male components were effective pollinators. The highest yield in large-scale seed production for ZP 427 was achieved in 2016, reaching 6,819 kg/ha on an area of 96 ha. For ZP 606, the highest seed yield was 5,526 kg/ha, recorded in 2015 on a 45 ha field. Testing has shown that the female components of these ZP hybrids are highly suitable for mechanized tassel removal using the Castrix machine, which significantly reduces labor requirements and lowers seed production costs. Due to these advantages, ZP hybrids are highly competitive in seed production, enabling collaboration exclusively with top-tier seed producers. The impact of unfavorable environmental conditions on seed production can be mitigated by establishing production on fertile soils with favorable structure, preferably on plots equipped with irrigation systems. Another important measure is to produce seed of the same hybrid at multiple locations, which is particularly beneficial in reducing the risks associated with weather extremes. Stable seed production is essential for maintaining and enhancing the market position of maize hybrids, both in Serbia and internationally.

Keywords: *maize; seed production; drought*

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Growth trend of organic maize production in Serbia

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Maize (*Zea mays* L., family *Poaceae*) is one of the world's most important crops. Organic farming is defined as an agricultural management system that supports and enhances natural biodiversity, utilizing processes and technologies based on biological principles without the use of artificial inputs or genetically modified organisms. One of the main prerequisites for successful organic corn production is the use of hybrids well adapted to local conditions and tolerant to disease and pest pathogens. The aim of this study was to show the range of areas under organic maize in Serbia for the period 2011 - 2023. Data were obtained from the Ministry of Agriculture, Forestry and Water Management of the Republic of Serbia. The following methods were used in the study: desk research, content analysis, comparative analyses and analyses of base and chain indices. In Serbia, organic grain production in 2023 covered an area of 5,372.23 hectares, with wheat occupying the largest area at a total of 2,010.33 ha, followed by rye with 844.76 ha, while maize, with a total of 603.93 ha, ranked fifth. During the analyzed period, the area under organic maize production varied significantly. The smallest production was recorded in 2011 (115.28 ha), while the largest was in 2015 (1,912.36 ha). The largest areas are located in the Vojvodina region. Given the growing global demand for organic maize, in Serbia, organic maize production is a relatively recent development and still occupies smaller areas, especially in comparison to conventional maize.

Keywords: *maize; areas; regions; base indices; chain indices*

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Global trends in bean production over the past decade

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Common bean (*Phaseolus vulgaris* L.) is one of the most important legumes in human nutrition worldwide, due to its rich nutritional profile and nitrogen-fixing ability. The aim of this study was to analyze global bean production from 2013 to 2022, with a focus on leading producers, regional trends, and potential for sustainable development. Data were obtained from the FAOSTAT database (Food and Agriculture Organization of the United Nations) and statistically processed using descriptive analysis and linear trend methods. The results show that total global bean production increased from approximately 26 million tonnes in 2013 to over 31 million tonnes in 2022. The largest producers during the analyzed period were India, Myanmar, Brazil, Mexico, and Tanzania. Asia dominated overall production, but a significant increase was also observed in African countries, highlighting the growing importance of beans as a source of food and income in developing regions. Most countries recorded yield growth, attributed to improved agronomic practices and variety selection. This analysis confirms the strategic role of beans in the global food system, particularly in the context of climate change and the need for sustainable agriculture. It is recommended to further improve local production systems, invest in the development of tolerant varieties, and support small-scale farmers to contribute to food security and poverty reduction in the world's most vulnerable regions.

Keywords: common bean; global production; FAO; agricultural

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Variability of seed maize inbred lines depending on the degree of defoliation

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The research examined the variability of maize inbred lines depending on different intensities of defoliation. Three maternal commercial inbred lines were tested in 2016 and 2017. at two locations (Zemun Polje and Parage). The experiment was set up in two sowing densities: 50,000 plants/ha and 65,000 plants/ha. Five different defoliation treatments were applied (T1- panicle removal, T2- the first leaf panicle removal, T3- the two apex leaf panicle removal, T4- the three apex leaf panicle removal, and T5- the removal of the panicle together with the four apical leaves. The following characteristics were examined: grain yield, number of rows of grains, number of grains in a row, cob length, weight of 1000 grains, germination energy, germination and fractional share in seed mass. Inbred line L217RfC showed the lowest value of yield variation, while line L335/99 showed the highest grain yield values in all ecological conditions, sowing densities and applied treatments. The results of statistical tests, with special reference to grain yield, indicate the importance of the intensity of defoliation in seed crops. A significant decrease in grain yield was found in inbred lines with removal of panicles with three leaves, and an even more significant decrease in yield was determined with treatment removal of panicles with four leaves. A high significance of ecological conditions (year × location) was established, where 2016. was a more favorable year, and the location of Parage had the more favorable agroecological conditions during both years of field experiments. The results of the research indicate a weak influence of sowing density on the yield and grain quality. Factors like: genotype, defoliation treatments and environmental conditions, as well as their mutual interactions, showed a high significance on the manifestation of grain yield and yield components by analysis of variance ANOVA.

Keywords: Maize; inbred lines; seed quality; yield; defoliation

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Maize variety verification and genetic purity methodologies: Theory vs. practice

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The basis of sustainable maize production relies on high-quality seed, with variety verification and genetic purity being essential determinants. Conventionally used grow-out tests (GOTs), while indispensable for assessing overall phenotypic expression under field conditions, are time-consuming, resource-intensive, and susceptible to environmental influences, often providing results too late in the seed production cycle for effective intervention. Biochemical markers (seed storage proteins/isozymes) in most cases environmentally independent, enable quick and precise analysis of maize varieties uniformity and purity. Still, biochemical assays cannot always effectively distinguish closely related inbred lines or hybrids. Advancements in molecular marker technologies have revolutionized genetic purity assessment. Simple Sequence Repeats (SSRs) and, increasingly, Single Nucleotide Polymorphisms (SNPs) offer highly precise, rapid, and environmentally independent means for distinguishing varieties and detecting off-types. DNA-based methods enable rapid and efficient analyses, significantly reducing time and land required, compared to traditional field trials. New emerging technologies such as hyperspectral imaging and machine learning algorithms are showing promise for non-destructive and automated seed identification based on miniscule phenotypic differences. Despite these technological leaps, practical integration of advanced methods includes challenges, such as initial investment in laboratory infrastructure, high costs of analytical consumables, the need for standardized protocols and their inclusion into national regulations. Furthermore, the volume and diversity of maize germplasm requires robust reference databases for accurate comparison. In conclusion, an integrated strategy combining the essential field-based validation of GOTs with the precision and speed of laboratory techniques, is vital. This should strengthen seed quality assurance systems, safeguard varietal integrity, and ultimately enhance maize productivity and food security globally, ensuring that farmers receive the precise genetic material they intend to plant.

Keywords: *maize, varietal integrity, grow-out test, advanced methods, integrated strategy*

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Maize inbred lines uniformity assessment using biological markers

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Monitoring the variability of agro-morphological traits is a valuable approach for assessing phenotypic uniformity and identifying traits specific to particular genotypes. Biochemical markers and Simple Sequence Repeat (SSR) markers are widely utilized for evaluating genetic uniformity and characterizing maize genotypes. In this study, a set of 15 newly developed maize inbred lines (G1–G15), originating from different breeding programs at the Maize Research Institute Zemun Polje, was selected for agro-morphological characterization and uniformity assessment using various marker types. Over a three-year field trial, agro-morphological traits—totaling 32 in accordance with UPOV descriptors—were evaluated, and phenotypic uniformity was assessed using the Off-type approach. Genetic uniformity was examined through Ultra Thin-Layer Isoelectric Focusing (UTLIEF) of reserve proteins (albumins and prolamins), complemented by analysis with eight SSR markers. Morphological markers revealed deviations from uniformity only in lines G1 and G2, a finding confirmed by biochemical and SSR analyses. Additionally, minimum and maximum average values for certain traits were observed in genotypes G3 and G11. Regarding UTLIEF results, genotype G14 displayed the lowest number of protein bands. The SSR analysis confirmed genetic divergence among the studied genotypes. Notably, unique alleles were identified in three lines: in G3 at the *umc 109275* locus (~141 bp), in G14 at the *umc 1448* locus (~161 bp), and in G11 at the *umc 1545* locus (~76 bp). These unique alleles can aid in genotype identification, elucidate genetic relationships and origins, and potentially be linked to important traits valuable for breeding programs. Based on the findings, 13 of the 15 inbred lines have achieved the required level of homozygosity, making them suitable candidates for hybrid seed production.

Keywords: *morphological markers; SSR markers; uniformity; UTLIEF; Zea mays L.*

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The impact of laboratory testing conditions on seed germination in different plant species

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The main purpose of standardizing conditions of testing seed germination is to ensure uniformity in the results which should be comparable to those obtained by other seed testing laboratories. Therefore, testing conditions and criteria are prescribed by national and international (ISTA) regulations. For each plant species, substrate, dormancy-breaking method(s) and testing duration are prescribed. The main goal of the germination test is to induce expression of the maximal germination potential and, in many cases several methods are recommended for a certain plant species. The seed testing laboratory of the Maize Research Institute Zemun Polje investigated the effect of substrate (for soybean), duration of pre-chilling, as the dormancy-breaking method (for barley and wheat), and the timing of evaluation on germination parameters (for maize). The effect of substrate on soybean seed germination indicated that the organic substrate induces higher percentage of seedlings with normally developed structure in comparison to the filter paper method. The duration of pre-chilling was investigated on the germination of cereal seeds by exposing samples for 3 or 6 days to low temperature (10°C). The obtained results indicate that the percentage of normally developed seedlings of barley was not affected, while wheat positively reacted to the longer exposure. The timing of evaluating germination energy (early germination) significantly affected the percentage of normal seedlings in maize inbred lines. The significantly higher percentage was recorded after a one-day delay (5th instead of 4th day). Due to the fact that germination energy provides valuable information on seed performance under field conditions, further practice in the laboratory was established to postpone evaluation of the germination energy in this specific case. Results obtained from this research suggest the necessity of selecting the most appropriate combination of germination testing conditions, which would provide induction of the maximal seed germination potential.

Keywords: *effect of substrate; pre-chilling; timing of evaluating germination energy*

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Germination evaluation criteria for the maize secondary root system

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Maize is the leading cereal crop in terms of worldwide production and trade, so it is of high importance to have uniform germination evaluation criteria. Although ISTA and AOSA rules have the same criterion for root system evaluation, the guidelines are not precise regarding the number and size of secondary roots that can replace defective primary roots. During normal growth and development, maize produces multiple root types, including primary, secondary, crown, brace roots, and associated lateral roots, to absorb water and nutrients. Carryover maize seed lots, which are commonly traded, especially in the case of inbred lines, more often contain seedlings with reduced and defective primary roots as a result of seed ageing. The objective of this study was to identify the minimum number and length of secondary roots to consider a maize seedling normal. Naturally aged seed was used in the experiment, i.e., seedlings that have naturally defective primary roots. Four hybrids created in the Maize Research Institute Zemun Polje of different types and maturity groups (FAO 200-700) were used for the study. Laboratory and field experiments were conducted. Plant/ear height, total leaf number, silking/tasseling emergence, yield components, and yield were observed traits. The findings revealed that seedlings with a defective primary root could still produce field-grown plants comparable to those with intact roots, provided they had at least two secondary roots of length equal to the coleoptile. Tasseling and silking were significantly different, but this was not reflected in the yield as the most important agricultural trait. Measured yield per ear was reduced when the primary root was defective, but this was not significant in the case of two secondary roots. This study offers valuable insights into the criteria for maize root system evaluation during germination, with implications for the seed trade. Further research encompassing a broader range of maize genotypes is recommended to refine these criteria and improve seed quality assessments.

Keywords: *maize; primary root; secondary roots; germination*

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The effect of aqueous ragweed (*Ambrosia artemisiifolia* L.) extracts on the germination and early growth of crops

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Ambrosia (*Ambrosia artemisiifolia*) is a widely distributed invasive species known for its pronounced allelopathic potential. Through allelochemicals released into its immediate surroundings, this species significantly inhibits the germination and early development of neighboring plants. Although its impact has been recognized, many aspects of its allelopathic mechanisms remain insufficiently researched, limiting a full understanding of its interaction with cultivated plants under natural and agricultural conditions. The aim of this study was to examine the effect of aqueous extracts of dried ambrosia leaves, in concentrations of 10%, 5%, 2.5%, and 1.25%, on the germination and early growth of six economically important agricultural species: barley (*Hordeum vulgare*), clover (*Trifolium repens*), pea (*Pisum sativum*), bean (*Phaseolus vulgaris*), corn (*Zea mays*), and wheat (*Triticum aestivum*). Seeds were germinated in a thermostat at a temperature of 26°C, with the control group treated with distilled water. Afterward, the tested species were grown under controlled laboratory conditions. The research methodology included the preparation of extracts according to a standard procedure, their application during a seven-day treatment period, and monitoring of parameters such as: germination percentage, mean germination time, germination index, as well as root and shoot length in the early developmental phase. The results showed that all tested extract concentrations had an inhibitory effect on germination and growth, with clover and pea being the most sensitive to allelopathic influence, while wheat and corn exhibited the highest degree of tolerance. Root and shoot growth analysis indicated a significant reduction in development, with inhibition in some cases exceeding 50%, particularly in pea and corn. Wheat proved to be the most resistant species to the allelopathic effect of ambrosia. The findings indicate the pronounced allelopathic activity of ambrosia and highlight the need for timely detection and control of its spread in agroecological conditions to preserve biodiversity and ensure stable agricultural production

Keywords: *Ambrosia artemisiifolia*; allelopathy; invasive species; germination; crop plants

Empowering traditional knowledge and biodiversity conservation through a citizen science approach

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Modern agricultural practices significantly reduce agro-biodiversity by controlling seed production and promoting new varieties with a narrow genetic base. This research employs citizen science as an innovative research approach that promotes close collaboration between scientific investigation and practical implementation. The aim of this collaborative initiative is to enhance understanding of local ecosystems while advocating for the conservation of their diversity. The network of scientists, volunteers, and farmers aims to strengthen and preserve traditional knowledge—an essential way for addressing biodiversity decline and the challenges of climate change. The research was conducted across the territory of Vojvodina using a random sampling design, targeting citizens in rural areas and employing the Q&A survey method to gather feedback and insights from respondents. Findings from volunteers' participation in research on the conservation of indigenous varieties reveal that only a small proportion of citizens are familiar with the terms “biodiversity” and “agro-biodiversity”. Additionally, only 6.1% maintain private collections of native varieties' seeds, although 84% express concern about climate change. In contrast, awareness and commitment regarding sustainable agriculture are notably higher, with 51.5% of those surveyed demonstrating understanding and involvement. These results suggest that factors contributing to biodiversity loss are not solely linked to modern agricultural practices but also include the erosion of traditional knowledge and skills within the population. Targeted educational initiatives aimed at younger generations could promote a deeper understanding of environmental issues and emphasize the importance of agro-biodiversity for ecosystem conservation. Moreover, preserving traditional knowledge and practices can lead to the development of sustainable scientific methodologies.

Keywords: *participatory science; indigenous seed; crop biodiversity*

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Climatic influence on the phenological development of cultivated *Allium ursinum* L. in the Mačva Region

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Allium ursinum L. (wild garlic) is a perennial herbaceous species belonging to the family *Amaryllidaceae*, widely recognized for its medicinal, nutritional, and culinary value. Owing to its rich phytochemical composition and increasing market demand, the interest in its systematic cultivation has been rising. However, the achievement of stable and economically viable yields remains constrained by pronounced climatic variability. This study aimed to assess the impact of climatic fluctuations on the phenological development of cultivated *A. ursinum*. The field trial was conducted in the agroecological conditions of the Šabac–Loznica region (44°39'41.8"N, 19°20'55.2"E), on loam soil, using vegetative propagation through bulb division in October 2020. Throughout five consecutive growing seasons (2021–2025), the progression of key phenophases emergence, vegetative growth, flowering, and senescence was monitored. Microclimatic parameters (air temperature and relative humidity) were recorded daily using automated data loggers, while precipitation data were sourced from the Republic Hydrometeorological Service of Serbia (RHMZS). The results indicated statistically significant interannual differences in phenophase duration and reproductive output (ANOVA, $p < 0.05$). Compared to the 30-year climatic baseline (1991–2020), February temperatures exceeded the average by up to 2.8 °C, whereas in April 2025 minimum temperatures dropped to –7 °C. Precipitation totals ranged from 15.8 to 73.6 mm, and relative humidity from 68% to 81%. Thermal and drought stress during the 2022 and 2024 seasons led to a reduction in seed capsule formation of up to 72%, while vegetative regeneration declined by 50%. Flowering and senescence were identified as the phenological stages most susceptible to climatic extremes. These findings emphasize the adverse effects of climate variability on the phenological performance and productivity of *A. ursinum*, underscoring the necessity for adaptive cultivation strategies under increasingly unpredictable environmental conditions.

Keywords: vegetative propagation; reproductive potential

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ECO-READY towards Consumer-Driven resilience for a sustainable European food system

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With its real-time surveillance system, the EU-funded ECO-READY project will enable that the challenges and interests of farmers and society at the European level are represented in future policymaking and monitoring. ECO-READY develops an Observatory (digital infrastructure) that brings together all existing data, models and knowledge on food security in the context of impact of the climate change on agro-biodiversity and invasive species and genetic resources, sustainable production, animal and plant diseases, and nutritional composition. With forecasts updated regularly and continuously, the observatory will offer real-time assessments for the food system along with policy recommendations, contingency plans, and resilience solutions created by 10 Living Labs. The data will be communicated via the digital observatory, informing citizens and supporting policy makers and agri-food supply chain stakeholders to make evidence-based decisions. In addition to the development of a digital observatory, ECO-READY is dedicated to scenario development (short- and long-term) activities that explore agricultural practices, policy changes, innovative farming ideas, and consumer-producer interactions to help mitigate future shocks. In order to determine their regional food priorities, the ten ECO-READY living labs covering the majority of Europe's geo-climatic zones have chosen important drivers. Taking into account drivers, for each of the living labs tailored-made scenarios relevant to their region and selected priority crops were created. ECO-READY, in its intention of solving the climate-food-biodiversity challenges, involved all relevant stakeholders for working together across different fields and taking real action that includes everyone and leads to lasting change.

Keywords: *scenario development; biodiversity; climate change; food security*

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Evaluation of the antioxidant response of maize to abiotic stress

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Maize (*Zea mays* L.), owing to its broad natural diversity, is an excellent source of phytochemicals, which play an important role in preventing various diseases. In the context of climate change, the increasing intensity and frequency of abiotic stress factors during the vegetation season may significantly alter the content of bioactive compounds in maize kernels. This study aimed to evaluate eight maize lines across two vegetation seasons for their content of antioxidants, i.e., free phenolic acids, carotenoids, and tocopherols. The analyzed compounds (p-coumaric acid (p-CoumA), ferulic acid (FA), cinnamic acid (CA), δ -tocopherol (δ -T), β + γ -tocopherol (β + γ -T), α -tocopherol (α -T), lutein+zeaxanthin (L+Z), and β -carotene (β -C)) were determined using high-performance liquid chromatography (HPLC) coupled with diode array (DAD) and fluorescence (FLD) detection. The results revealed a decreasing trend of FA concentration in all tested lines during the drier growth season. Similarly, reduced concentrations of p-CoumA and CA were observed under drought conditions in all tested lines, except in lines ZP7 and ZP4, which showed increased concentrations, indicating a better resilience to abiotic stress. Interestingly, most lines showed higher concentrations of δ -T, β + γ -T, and β -C during the drought year. In both growth seasons, the tested maize lines showed different patterns of increase and decrease in α -T and L+Z concentrations, indicating the differential activation of defense mechanisms against abiotic stress due to their varied genetic backgrounds. According to the obtained results, line ZP7, among all tested maize lines, can be considered as a promising source of bioactive compounds, maintaining high levels even under drought stress. Results from this study highlight the importance of identifying and selecting maize lines that possess both high nutritional value and resilience to abiotic stress, which is necessary due to ongoing climate change.

Keywords: *phytochemicals; drought tolerance; genetic diversity; secondary metabolites; maize breeding*

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Quality of whole grain bread and benefits in human nutrition

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The key role in determining the technological quality of bread is played by the content of gluten, which consists of the components gliadin and glutenin. Gliadins and glutenins determine the properties of elasticity and viscosity of the dough, which are important in the process of dough fermentation and the quality of the bread center that is ultimately formed during baking. Gluten proteins are very important for oriental bread (made from wheat flour, water, and yeast). Besides technological quality, bread is characterized by nutritional quality determined by the content of proteins (13%), carbohydrates (68%), fats (2%), minerals (2%), and vitamins, particularly found in the grain's outer layers and germ. This study reviews the quality and consumption benefits of whole grain bread. Micronutrient malnutrition, affecting 40% of the global population, particularly in developing countries, emphasizes the importance of whole grains. In 2024, over 2.2 billion individuals were micronutrient malnourished, indicating inadequate food security for essential nutrients needed by high-risk groups. Deficiencies in nutrients such as iron, iodine, zinc, selenium, calcium, and magnesium contribute to diseases and increased mortality. These deficiencies are pronounced in regions with limited access to fruits, vegetables, and animal products, notably in underdeveloped Asia and sub-Saharan Africa, with up to 20% of the European population also deficient in minerals. Micronutrient deficiencies, a significant global health challenge, notably affect vegetarians and impair children's cognitive abilities, educational achievements, and labor productivity. Whole grains, minerals, and antioxidants are associated with a lower risk of heart disease and colon cancer, help lower LDL cholesterol, improve endothelial function, and reduce blood pressure and type 2 diabetes, enhance cognitive function, and prevent inflammatory or chronic diseases. The World Health Organization (WHO), Dietary Guidelines for Americans, and European Food Safety Authority (EFSA) advocate that at least half of grain intake should consist of whole grains, with a fiber intake recommendation of 25-38 g day⁻¹. The mineral content of whole grain flours varies based on wheat genotypes and agricultural practices. Breeding efforts should focus on developing wheat varieties that offer better micronutrient bioavailability and higher antioxidant levels.

Keywords: bread; gluten; wheat seed; minerals; quality

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Status of the dietary fibre and phenolic compounds in some standard and ancient cereals

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The popularity of the functional food grows with the increasing awareness of consumers of its health promoting characteristic. The cereals with various active compounds are of particular importance. This research aimed to analyse whole-grain cereals (bread wheat, durum wheat, triticale, rye, barley and oat), and some ancient grains (emmer wheat and spelt) towards bioactive compounds, such as dietary fibre (arabinoxylan and β -glucan), and specific phenolic compounds. Genotypes of durum wheat, triticale, spelt, emmer wheat and barley could be considered as important and sustainable sources of prebiotic fibre (β -glucan and arabinoxylan, ranging 0.11–4.59% and 0.51–6.47%, respectively). The presence of various phenolic substances was recorded in genotypes of the examined cereals. The greatest concentration of p-coumaric acid and ferulic acid was mostly determined in Caramel oat, while the triticale (Agrounija) was highest in dihydrocaffeic acid. Dihydro-p-coumaric acid, naringin, quercetin, epicatechin were determined in grains of oat (Sopot), while catechin was present in barley grains (Apolon and Osvit) underlining their unique chemical profile. The naringenin was found in the grains of Emmer LP2-1-5 and oat genotypes. This research provides valuable information of specific nutritional profile of cereals, indicating their importance as nutraceuticals. It also provides genetic background that could be translated to genotypes with even more profound effects on human health.

Keywords: *functional food; phenolics; dietary fibre*

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Soybean hulls as a sustainable source of bioactive compounds: Unlocking functional and health-promoting potential

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Soybean hulls, an abundant by-product of soybean processing, have traditionally been underutilized or relegated to low-value applications such as animal feed. However, emerging evidence suggests that soybean hulls are a promising source of bioactive compounds with significant functional and health-promoting properties. These include dietary fibers, phenolic compounds, flavonoids, and pectin-like polysaccharides, many of which exhibit antioxidant, anti-inflammatory, and potential prebiotic activities. The structural complexity and composition of the hull matrix offer a unique opportunity for the valorization of this agricultural residue through green extraction strategies such as enzymatic treatment, ultrasound-assisted extraction, and subcritical water processing. Recent studies have demonstrated that thermal, chemical, or enzymatic treatments can significantly enhance the release and functionality of these bioactives. Moreover, the integration of soybean hull-derived components into food systems and nutraceutical formulations may contribute to improved techno-functional properties, such as emulsifying capacity, water- and oil-holding capacities, and stabilization of food matrices. In addition, their antioxidant potential makes them valuable in oxidative stress-related health applications and food preservation. The current trend toward sustainable and circular bioeconomy further underscores the importance of re-evaluating agro-industrial by-products such as soybean hulls. Harnessing their bioactive potential not only reduces waste and adds economic value but also aligns with clean-label and functional food innovations. This review aims to highlight the compositional richness, bioactivity, and potential applications of soybean hulls, positioning them as a novel and sustainable source of health-promoting compounds in the food and pharmaceutical industries.

Keywords: *soybean hulls; bioactive compounds; antioxidant activity; sustainable utilization*

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Influence of genotype and environment on the fatty acid profile and α -tocopherol content in safflower (*Carthamus tinctorius* L.) oil

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Safflower (*Carthamus tinctorius* L.) oil is appreciated for its high nutritional quality, primarily due to a favorable fatty acid composition and the presence of bioactive compounds such as tocopherols, which contribute to oxidative stability and health benefits. With increasing interest in plant-based oils with functional properties, understanding the impact of genetic and environmental factors on oil composition is key to optimizing quality traits. This study examined the content of selected fatty acids—palmitic (C16:0), stearic (C18:0), oleic (C18:1), linoleic (C18:2), and linolenic acid (C18:3) as well as α -tocopherol in oil extracted from two safflower genotypes cultivated at three locations in Serbia (Pančevo, Rimski Šančevi, and Sombor). The oil was obtained by cold hydraulic pressing and analyzed using gas chromatography and high-performance liquid chromatography. The average contents of palmitic, stearic, oleic, linoleic, and linolenic acids were 6.77%, 3.29%, 13.35%, 75.32%, and 0.26%, respectively, while α -tocopherol averaged 456.66 mg/kg. ANOVA showed that palmitic acid, oleic acid, and α -tocopherol were significantly affected by genotype, location, and their interaction ($p < 0.001$), indicating high variability and potential for targeted improvement. Stearic acid was influenced by location and the genotype $\times$ location interaction ($p < 0.05$), but not genotype alone. Linoleic acid was affected by location and interaction, but not by genotype ($p = 0.95$). Linolenic acid was influenced only by location ($p = 0.04$), suggesting greater genetic stability. These findings highlight the importance of selecting optimal genotype–environment combinations to improve the nutritional and functional quality of safflower oil. Traits such as oleic acid and α -tocopherol, which are highly responsive to both genotype and environment, are promising targets in breeding programs. Meanwhile, the relative consistency of stearic, linoleic, and linolenic acids supports their value in achieving stable oil composition.

Keywords: safflower oil; fatty acid composition; α -tocopherol; nutritional quality

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Sustainable trends in maize cultivation: organic cropping system for improved bioactive profile of added-value ZP hybrids

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Organic maize production contributes to ecological sustainability and offers numerous benefits across the food supply chain. In recent decades, global interest in organic food has grown, and maize (*Zea mays* L.), as a major staple crop, plays a key role in this trend due to its nutritional importance and extensive use. This study investigated the impact of conventional and organic cropping systems on the bioactive profile of four added-value maize hybrids developed at the Maize Research Institute in Zemun Polje, Serbia. Compared to conventional systems, organic cropping systems resulted in significantly higher levels of total phenolic compounds, phenolic acids, and antioxidant activity, with mean increases of 14.45%, 8.99%, and 16.15%, respectively. These results suggest that organically produced maize has enhanced functional quality and potential health benefits. On average, the content of total phenols in maize hybrids in conventional cropping was 2527.16 µg GAE/g, while the highest content was recorded in maize hybrid ZP555 in an organic cropping system and amounted to 3296.57 µg GAE/g. The content of ferulic acid in maize genotypes ZP552b, ZPQPM13, and ZP555 was 7.03%, 5.31%, and 17.01% higher in organic than in conventional cropping systems, respectively. Although it can be said that the organic cropping system contributed to the improvement in antioxidant capacity, our results showed that the impact of genotype was predominant. The maize genotype ZP 5048 red is the most obvious example of this, exhibiting a contrary trend with decreased amounts of bioactive compounds and antioxidant activity under the organic cropping system. These findings suggest that, while organic cropping systems can enhance the nutritional and functional quality of maize, the selection of genotypes remains critical for ensuring the quality and sustainability of the final product. Further research is needed to investigate system-genotype interactions across a wider range of genotypes and environmental conditions.

Keywords: *organic cropping system; maize; bioactive profile; phenolic compounds; antioxidant capacity*

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Biscuit quality under acrylamide-targeted agronomy: A bilateral assessment

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The study aimed to evaluate the quality of biscuits prepared from cereal raw materials obtained through different agronomic practices designed to minimize the levels of acrylamide precursors prior to baking. A bilateral collaboration between Slovakia and Serbia enabled a comprehensive assessment of sensory attributes and texture characteristics using standardized sensory analysis and instrumental texture profiling. The experimental biscuits were evaluated by trained panels in both countries, focusing on key parameters such as appearance, aroma, taste, mouthfeel, and textural properties including hardness and fracturability. The results demonstrated that the agronomic interventions had no significant impact on the overall sensory quality or texture of the final products. These findings suggest that such cultivation strategies can be implemented to reduce potential health risks without compromising consumer-acceptable biscuit quality. This international approach reinforces the value of cross-border research in addressing food safety while preserving product quality.

Keywords: *biscuits; sensory analysis; textural analysis; acrylamide*

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Free asparagine and sugars in various maize genotypes as principal precursors of acrylamide formation in food

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Maize is used in the production of a wide range of foodstuff, including whole-grain flour, semolina, extruded snacks, and bakery products, making maize-based foods one of the main dietary sources of Maillard reaction products. Free amino acids (*free asparagine*) and reducing sugars are considered the primary precursors in the formation of heat-induced contaminants such as acrylamide. In order to identify genetic resources with reduced potential for acrylamide formation, the contents of free asparagine and reducing sugars were analyzed in a nineteen maize genotypes cultivated during the 2021, 2022, and 2023 growing seasons at the Maize Research Institute, Zemun Polje. HPLC was used to analyze free asparagine with a DAD detector, while sugars have been analyzed with a RID detector. The obtained results show that free asparagine and reducing sugar content varies between maize genotypes. Our findings show no statistically significant difference between the three years in terms of free asparagine content. However, as it was established, the impact of the environmental conditions was reflected in the sugar content. Principal Component Analysis (PCA) was performed to evaluate genotype differences over the years. Three groups of genotypes can be distinguished according free asparagine content varying from 200 µg/kg to more than 500 µg/kg. The average content of fructose, glucose, sucrose, and maltose was approximately 55%, 54%, 7%, and 42% lower in genotypes cultivated in 2022, and 3%, 41%, 12%, and 16% lower in those cultivated in 2023, compared to 2021, respectively. These results suggest that both genetic background and environmental factors, particularly year-to-year variation, have significant effects on the content of acrylamide precursors in maize, emphasizing the importance of genotype selection for safer maize-based food production.

Keywords: *free asparagine; sugars; precursors; acrylamide; maize*

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Blue maize as source of anthocyanins with diverse pharmacological activities: Microencapsulation and fate during accelerated aging and in vitro gastrointestinal digestion

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Among other plant species, cereal grains can also be a source of anthocyanins (ACNs). Whole-grain flour of colored maize has primarily found application in the production of bakery and confectionery foodstuffs. However, application of ACNs originating from maize grains in pharmacy requires their isolation and represent a certain challenge. The aim of the research was to develop microencapsulation systems of ACNs from waste product of blue maize processing using maltodextrin (MD) and hydroxypropyl- β -cyclodextrin (HPCD). Liquid blue maize extract was spray dried with and without adding carrier agents: MD (30%), HPCD (30%) and combination of both carriers (15% MD and 15% HPCD). The obtained samples were analyzed for the content of ACNs before and after accelerated aging, as well as for the fate of ACNs in the digestive system. Total ACNs content in the spray dried maize extract (SME) was 12846 mg CGE/kg, with acylated Cy-3-(6'-Mal-Glu) than Cy-3-Glu as dominant. Microencapsulates had a lower content of total anthocyanins by 16, 14 and 26% compared to SME, respectively. Cavity and hollow molecular shape of the CDs make them ideal carrier to decrease phenolic compounds loss. So, after aging, the content of total ACNs detected by HPLC was 11% lower in microencapsulates with MD, compared to that before aging, while in microencapsulates with HPCD the difference was not statistically significant. Due to the action of digestive enzymes, in the first and last stage of digestion of SME and microencapsulates with MD, HPCD and MD+HPCD, the content of ACNs in the fluids was 59, 66, 67 and 64% i.e. 49, 42, 57 and 56%, respectively, of their initial value. Waste product of blue maize processing in form of microencapsulates can contribute significantly to the daily intake of ACNs, especially acylated forms whose daily intake has been estimated at 23% compared to 77% of non-acylated ones.

Keywords: blue maize; processing waste; anthocyanins; microencapsulation; fate during aging and digestion

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Filter strip as a method for detection of antioxidative components in complex matrix: A case study in maize root apoplast

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The root apoplast, as a thermodynamically open system in direct contact with the environment, is the first to encounter varying environmental conditions that determine the response of the whole plant. In rhizosphere research, different techniques have been developed for the collection of root apoplast. The most commonly applied infiltration and/or centrifugation technique has serious disadvantages which significantly increase the risk of contamination with cytosolic or vacuolar components (the infiltration of solution into the plant tissue inevitably alters the chemical equilibrium existing in the apoplast, the tissue excision results in unavoidable contamination from injured cells, the applied centrifugal force causes membrane rupture and cellular injury). In order to overcome these drawbacks, we used filter paper strips to collect root apoplastic fluid (AF). This sampling technique, based on sorption media placed onto the root surface, is a non-invasive method for root AF collection, which allows experiments with intact plants. It is suitable for monitoring changes of metabolic compounds of the root apoplast, especially in hydroponically grown plants, avoiding mechanical stress, simultaneously providing adequate quantities of the apoplastic fluid for analysis. Our results demonstrate that the AF samples did not show the presence of intracellular components, indicating that these isolates were not contaminated with cytoplasmic constituents at all. Due to the low amount of starting material, the sensitivity of the assays was verified for antioxidative enzymes (SOD, POD), including isoelectrofocusing for their visualization, as well as metabolites (organic acids, sugars, and phenolic compounds). Dominant organic acid identified in AF was succinic acid, sugar glucose and phenolic compound coniferyl alcohol. To conclude, our results unequivocally demonstrate the advantage of using filter strips for collection of the apoplastic fluid from the intact plant roots, giving sufficient amounts of uncontaminated fluid containing enzymes and metabolites, such as organic acids and phenolics.

Keywords: root apoplast; filter strip; HPLC; antioxidative enzymes; isoelectrofocusing

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Antioxidant capacity and bioactive compounds of maize silk from different ZP genotypes for application in health-promoting dietary supplements

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Natural plant-based products have recently been growing in popularity in pharmaceutical and functional food industries due to their antioxidant properties. For centuries, maize silk (*Stigma maydis*) has been utilized in traditional medicine for the prevention and treatment of numerous medical conditions. The therapeutic, anti-inflammatory, antimicrobial, diuretic, antidiabetic, and antioxidant properties originate from silk's chemical composition, rich in polyphenols, polysaccharides, protein, fiber, alkaloids, vitamins and minerals. Maize silk of five ZP maize genotypes (ZP 355su, ZP 6119k, ZP 5550, ZP 666 and ZP 6263) harvested at three different maturity stages, i.e., days of silking (DS), was used to identify the most promising raw material for the production of health-promoting dietary supplements. The study focused on investigating the contents of bioactive compounds and antioxidant capacity of maize silk. Antioxidant capacity was evaluated after following ABTS⁺, DPPH[•], IC₅₀, and FRAP methodologies for comparison. The genotype ZP 5550 (15 DS) exhibited the highest antioxidant potential. The most abundant bioactive compounds were free phenolic. Notably, genotype ZP 5550 (DS 15), has a significantly higher concentration of total free phenolics, approximately six time higher than those observed in genotype 6119k (15 DS). The total protein content ranged from 12.02% in genotype ZP 6263 (15 DS) to 19.43% in genotype ZP 6119k (7 DS). The findings of this study may have significant implications for maize breeding initiatives and the identification of the most promising hybrids for the manufacturing of functional foods and nutritional supplements.

Keywords: *maize silk; maturity stage; bioactive compounds; antioxidant capacity; traditional medicine*

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Chemical composition of tobacco seed (*Nicotiana tabacum* L.)

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The chemical composition of tobacco seed (*Nicotiana tabacum* L.) varies significantly depending on the genetic characteristics of the variety and environmental conditions during its development. This paper presents a review of the chemical composition of tobacco seed based on relevant scientific literature. The average composition (g/100 g dry matter - DM) includes: moisture 5.88 g, total ash 3.20 g, total fats 34.58 g, total proteins 26.29 g, and carbohydrates 30.06 g. The results show that fats are the most abundant component, with linolenic acid (45.1 g/100 g DM), oleic acid (26.5 g/100 g DM), and palmitic acid (18.0 g/100 g DM) being the dominant fatty acids. The oil content in the seed ranges from 36% to 41%, depending on the variety and agroecological conditions. Among mineral elements, calcium (Ca) and potassium (K) are the most prevalent. It is important to note that dry tobacco seed does not contain nicotine, as its biosynthesis begins only during the germination process. After harvest, the seed contains about 16% moisture, which complicates storage. To prevent spoilage and preserve germination potential, moisture content must be reduced below 7%, which can be achieved through natural sun drying with ventilation or by using drying chambers under controlled conditions. The obtained data may serve as a basis for further research on the nutritional and physiological properties of tobacco seed, as well as for optimizing its storage and utilization in various industrial sectors.

Keywords: tobacco; seed; chemical composition; fatty acids; nicotine

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Valorization of Pea Pod waste and seeds for the development of novel food products

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The *Pisum sativum* L. (green pea) is one of the most widely consumed legumes, or vegetable crops, known for fresh eating, processing, or livestock feed. Besides the edible part of the pea, 35%-40% consists of solid waste (pods), an abundant by-product with health-promoting properties. Although pea pods are often thrown away, this waste contains many bioactive compounds (polyphenols and natural pigments). Pea powder can serve as an encapsulation carrier, efficiently using both pea pods and seeds. This study aimed to optimize ultrasound-assisted extraction (UAE) to increase the yield of bioactives from pea pods and to encapsulate the extract, maximizing the value of both pea components. Extraction parameters were varied, including time (15 min, 30 min, 1 h), ethanol concentration (30%, 50%, 70%), and the solvent-to-sample ratio (1:10, 1:20, 1:50), using Design Expert software. The best pea pod extract (58.51% ethanol, extraction time of 45 min, and a solvent-to-sample ratio of 1:48) was concentrated and encapsulated in three forms: pure pea pod extract (1); extract combined with freeze-dried pea powder (2); and combined with whey protein (3), a common protein carrier. Total polyphenolics (TPC) and natural pigments (chlorophyll a, chlorophyll b, and carotenoids) were analyzed in each sample to assess how different carriers affected compound retention. UAE efficiently extracts high levels of bioactives from pea pods, enabling uses across various industries. All encapsulates had high levels of bioactives, although adding freeze-dried pea powder significantly increased chlorophyll a and b to 1758.40 µg/g and 2539.86 µg/g. Using pea powder as an innovative carrier for stabilizing bioactives from pea waste highlights the potential of both as nutraceuticals and functional foods. Both peas and pea pods can be used as key nutrients to support healthy diets, valorize this by-product as an alternative food, and promote sustainable agricultural waste management.

Keywords: *pea pod waste; seed, innovative carrier; encapsulation; ultrasound-assisted extraction*

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Innovative strategies for valorizing oilseed cakes in sustainable food systems

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Oilseed cakes, the nutrient-rich by-products generated during oil extraction, are gaining increased attention as valuable resources within sustainable food systems. This study highlights two main strategies for developing innovative food products: the use of novel, alternative raw materials not traditionally incorporated in food production, and the valorization of by-products such as oilseed cakes. For these approaches to succeed, coordinated efforts involving updated regulations, consumer education, and transparent labeling are essential to foster consumer acceptance and trust. Oilseed cakes present multiple benefits, including waste reduction, low-cost production of nutrient-dense ingredients, and alignment with circular economy principles where “waste = food.” Their generally good shelf stability is a notable advantage, although improper storage can lead to rancidity and off-flavors. Research indicates that moisture activity in oilseed cakes remains stable for up to four months, effectively limiting microbial growth. Additionally, storage in environmentally friendly paper packaging preserves sensory and nutritional qualities for up to six months at room temperature. Traditionally, oilseed cakes have been used primarily as animal feed, biofuel substrates, or disposed of in landfills, where decomposition poses environmental risks such as methane emissions and water contamination. However, modern valorization techniques focus on extracting high-value functional ingredients like protein isolates, antioxidants, dietary fibers, enzymes, and bioactive compounds, which have applications across food, cosmetics, agriculture, and pharmaceutical industries. Emerging innovations include developing functional food ingredients, biopolymer packaging, enzyme production via fermentation, and substrates for medicinal mushroom cultivation. By transforming oilseed cakes into value-added products, these strategies not only contribute to environmental sustainability but also support food security and circular economy goals, positioning oilseed cakes as versatile resources within sustainable food systems.

Keywords: *oilseed cakes; by-product valorization; food industry; functional ingredients*

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Advances in sunflower oilcakes for food industry applications

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Sunflower oilcake, a by-product remaining after oil extraction via screw pressing, has traditionally been used as animal feed. However, growing global demand for sustainable protein sources highlights its potential as a human food ingredient. Animal proteins are costly and resource-intensive, while many plant proteins have limitations in taste, functionality, and price. Sunflower oilcake proteins, currently containing about 30% protein with limited functional properties, can be improved through hull removal to yield protein concentrates with increased protein content, enhanced digestibility, lighter color, and better functional characteristics. The composition of sunflower oilcake depends on sunflower hybrids and oil extraction methods, with protein and fiber content varying significantly. Residual oil content is typically well below 3% when solvent extraction is used, whereas mechanical pressing usually leaves a residual oil content well above 7%. Phenolic compounds, mainly chlorogenic and caffeic acids, contribute to nutritional value but can affect protein functionality through Maillard reactions and protein-polyphenol complex formation, potentially reducing lysine content and digestibility. Despite lysine being a limiting amino acid, sunflower proteins can complement other protein sources within a balanced diet. Beyond feed, sunflower oilcake contains valuable bioactive compounds like antioxidants, fibers, vitamins, and minerals, which can be extracted for food, cosmetic, or pharmaceutical use. Functional sunflower proteins find applications in emulsions, baked goods, meat and milk substitutes, and protein bars. Their allergen-free, non-GMO nature, affordability, and sensory qualities make them attractive for diverse food products. This review highlights sunflower oilcake valorization for sustainable food production, emphasizing improved protein extraction, functional properties, and broad industrial applications, supporting a circular bioeconomy.

Keywords: *sunflower cake; food applications; functional properties; bioactive compounds*

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A green biotechnological approach to enhancing *Cotinus coggygia* leaf extracts via lactic acid fermentation

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The smoke tree (*Cotinus coggygia* Scop., Anacardiaceae) is a traditional medicinal plant known for its wide range of pharmacological activities, including antioxidant, anti-inflammatory, and antimicrobial effects. This study investigates, for the first time, the effect of spontaneous lactic acid fermentation on the extraction efficiency and phytochemical composition of ethanolic leaf extracts, using high-performance thin-layer chromatography (HPTLC). Ethanolic extracts of non-fermented (NF) and fermented (F) samples were compared based on their chromatographic fingerprinting profiles using various detection methods: 0.5% natural product (NP) reagent (w/v) and 5% polyethylene glycol (PEG, w/v), AlCl₃, and FeCl₃ for phenolic profiling, p-anisaldehyde reagent for terpenoid profiling, and HPTLC-DPPH bioautographic assay. Optimal separation of all compound classes was achieved using a mobile phase composed of hexane: ethyl acetate: and acetic acid (15:9:1, v/v/v). In the F smoke tree sample, derivatization with NP/PEG and FeCl₃ revealed an intensified and more complex phenolic profile compared to NF samples, while AlCl₃ derivatization confirmed the enhanced presence of flavonoids. These findings suggest that fermentation facilitates the release of bioactive phenolic and flavonoid compounds by breaking down plant cell structures and glycosidic bonds. Immersion of the chromatogram in DPPH solution revealed antioxidant activity in the ethanol extract of the F sample, indicating enhanced extractability of phenolic compounds due to fermentation. Fermentation increased the extraction of phenolics by 45%. A notable increase in terpenoid zones was observed in fermented extracts, accompanied by significantly enhanced antioxidant activity. Total phenolic content and flavonoid content were found to be strongly correlated ($r^2 = 0.95$). Additionally, there was a positive correlation ($r^2=0.79$) between flavonoids and antioxidant activity. The application of *Lactobacillus* fermentation enhances the levels of phenols, flavonoids, and terpenes in smoke tree extracts, yielding a sustainable and low-cost biotechnological approach that offers new opportunities for developing natural products with improved bioactivity.

Keywords: *Cotinus coggygia* Scop.; *Lactobacillus* fermentation; green extraction; HPTLC fingerprints; antioxidant activity

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Simultaneous ultrasound-assisted extraction of swertiamarin and isogentisin from *Gentiana lutea* L. leaves: Optimization by response surface methodology and storage stability

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Leaves of *Gentiana lutea* L., traditionally used for treating heart disorders, offer a sustainable and underutilized source of bitter secoiridoids and xanthenes, otherwise derived from *Gentianae radix*. As *G. lutea* is an overexploited species, the use of its more readily available leaves could help reduce the pressure on scarce resources caused by root harvesting. This study aimed to optimize the ultrasound-assisted extraction (UAE) of the secoiridoid swertiamarin and the xanthone isogentisin from *G. lutea* leaves using response surface methodology (RSM). The influence of extraction time (A: 5–65 min), ethanol concentration (B: 10–90% v/v), liquid-to-solid ratio (C: 10–50 mL/g), and temperature (D: 20–80°C) was analyzed at five levels according to a central composite design (CCD). The calculated optimal extraction conditions for the simultaneous maximization of swertiamarin and isogentisin yields were: A – 50 min, B – 30% v/v, C – 30 mL/g, and D – 62.7°C. Under these conditions, the experimentally obtained yields were 3.75 mg/g DW for swertiamarin and 1.57 mg/g DW for isogentisin, closely matching the RSM model predictions. The stability study revealed that low-temperature storage preserved major bioactive compounds, whereas mangiferin stability was compromised by elevated temperature and light exposure. The established models support the production of standardized *G. lutea* leaf extracts and may facilitate the efficient separation and purification of their bioactive compounds, thereby contributing to the further valorization of this valuable plant material.

Keywords: yellow gentian leaves; secoiridoids, xanthenes; ultrasound-assisted extraction; storage stability

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Supercritical CO₂ pretreatment of buckwheat for improved extraction of bioactive constituents

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Diversifying food resources through the incorporation of crops with nutritional and health benefits present a promising strategy in sustaining nutraceutical security under climate change and biodiversity degradation. Buckwheat is such underutilised crop, known for the abundance of bioactive compounds and nutrients that have a vast potential to support human health. Given the biological potential closely associated with polyphenolic content, particularly rutin, this study aimed to optimize ultrasound-assisted extraction (UAE) from the aerial parts of *Fagopyrum esculentum*. To assess the impact of raw material pretreatment, supercritical CO₂ extraction was initially applied. The resulting defatted biomass was then subjected to UAE, with response surface methodology (RSM) employed to evaluate key extraction parameters, including amplitude, pulse, and extraction time. All three studied parameters showed significant effects on the extraction efficiency of total phenolic compounds and rutin. Notably, pretreatment with supercritical CO₂ led to a marked increase in phenolic recovery compared to untreated biomass. According to RSM analysis, the treated material achieved optimal yields of 49.13 mg GAE/g DW of total polyphenols and 41.73 mg/g DW of rutin under conditions of 98.5% amplitude, 82.22% pulse, and 9.99 minutes. In contrast, under similar conditions (97.24% amplitude, 91.92% pulse, and 9.89 minutes), the untreated material yielded lower concentrations: 42.66 mg GAE/g DW of total polyphenols and 38.01 mg/g DW of rutin. Therefore, it can be concluded that supercritical CO₂ pretreatment has a beneficial effect on the downstream processing of buckwheat biomass, notably enhancing the efficiency of subsequent UAE. Processing the raw material derived from underutilised crops such as buckwheat holds significant potential to improve consumer nutrition and health and pave the way for incorporation into new natural products.

Keywords: buckwheat; supercritical CO₂ extraction; ultrasound-assisted extraction; response surface methodology; rutin

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Sustainable natural food colorant from beetroot waste: NADES-based extracts with promising functional properties

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Synthetic food dyes are associated with adverse health effects, including potential carcinogenicity and hyperactivity, which necessitate safer, natural alternatives. Natural pigments derived from agro-industrial waste not only provide vibrant coloration but also offer bioactive properties with potential health benefits. This study explores the valorization of beetroot waste through the extraction of bioactive pigments using 15 different Natural Deep Eutectic Solvents (NADES), compared to conventional solvents. Optimization of extraction conditions enabled evaluation of betalain content, thermal stability, total phenolic compounds, antioxidant capacity, and antimicrobial activity. Further assessments included anti-obesity and anti-diabetic potential, inhibition of enzymatic browning, and cytotoxicity against Caco-2 cells to identify the most bioactive extracts. NADES 6 and 15 yielded extracts with superior bioactivity, demonstrating their efficacy as sustainable solvents. As a proof of concept, the five most potent extracts were applied as natural colorants in cookie formulations, exhibiting both chromatic stability and organoleptic properties. This study explores a circular economy strategy to create natural, health-promoting food colorants from underutilized agricultural byproducts, in line with global trends favoring sustainability and clean-label solutions.

Keywords: beetroot waste; betalains; Natural Deep Eutectic Solvents (NADES); natural colorants; biological activities

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Fermentation Potential of Field Pea (*Pisum Sativum*): A Nutritional Perspective

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The aim of this study was to determine the physicochemical properties of five physiologically ripe *Pisum sativum* (pea) varieties – “Dunav”, “Tamiš”, “Kelvedon”, “Mali Provansalac” and “Čudo Amerike”- in order to evaluate their potential as substrates in the fermentation process. The analysis focused on key nutritional parameters with critical influence in microbial growth, fermentation kinetics, and development of desirable sensory and nutritional profiles in fermented products. Pea seeds, obtained from the Institute of Field and Vegetable Crops in Novi Sad, were milled into fine flour using a laboratory mill and stored at 4°C until analysis. Each variety was analyzed in duplicate, and mean values calculated per dry weight were used for comparison followed by statistical analysis. Standard laboratory methods were employed to quantify macronutrient composition. Results showed considerable variations among the five varieties, with protein content ranging from 21.14% (“Dunav”) to 26.81% (“Mali Provansalac”) and total sugar content from 1.25% (“Dunav”) to 4.20% (“Tamiš”). Beyond simple sugars, total carbohydrate content - including starch and dietary fiber - also varied among the samples. Fat content was relatively low across all samples (1.19% - 1.99%), consistent with typical legume profiles. Based on a comparative assessment, two varieties, one with the highest fermentable sugar content and balanced carbohydrate profile (“Tamiš”) and the other with the lowest sugar and comparatively lower protein levels (“Dunav”), were selected for further fermentation trials. Both varieties were selected for additional studies to evaluate how compositional extremes influence microbial activity and product quality in fermentation process. These findings provide a foundation for optimizing legume-based fermentation systems and support the development of sustainable, plant-based fermentation substrates with enhanced nutritional and functional value.

Keywords: *Pisum sativum*; nutritional analysis; protein; sugar; fermentation

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Maize breeding - challenges and needs

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Maize is generally well-suited to the climate of South-eastern Europe, but rising temperatures and lack of precipitation negatively affect yields. Breeding of new, climate resilient hybrids, came out as a main adaptation strategy. However, developing of materials capable to cope with a climate change is a complex task. While grain yield and moisture content remain dominant traits, there's a growing emphasis on heat and drought tolerance. Due to the increasing frequency and intensity of storms resulting from climate change, enhancing crop tolerance to stalk and root lodging is a crucial priority. An old idea, breeding for short stature maize, could be an interesting solution. Short-stature maize can offer increased tolerance to lodging, allowing for higher planting densities and potentially improved yields. These changes could lead to reduced risks associated with extreme weather, and more sustainable farming practices. Climate change is not just about the change of meteorological events. It cause change in biotic factors too (diseases, pests, weeds), making breeding for successful hybrids quite uncertain. A special attention should be paid in breeding for tolerance to mycotoxigenic species, such are *Fusarium graminearum* and *Aspergillus flavus*. Rising temperatures are predicted to lead to the emergence of more aggressive strains and to cause a shift in the prevalence of fungal species. In the last decade, there is a growing interest in developing maize hybrids with enhanced nutritional value and health benefits. A special attention is paid to pigmented maize, rich in anthocyanins, water-soluble pigments known for their anti-oxidative, anti-inflammatory, anti-bacterial, even anti-carcinogenic properties. Combining appropriate maize genetic diversity with modern breeding techniques is crucial for developing resilient and high-yielding hybrids, with elevated content of nutrients and bioactive compounds. This approach is especially important in the face of climate change and growing food demands.

Keywords: *maize; breeding; climate change; adaptation; tolerance*

Acknowledgment: This work is supported by the Centre of Excellence for Innovations in Breeding of Climate-Resilient Crops - Climate Crops, Institute of Field and Vegetable Crops, Novi Sad, Serbia.

Genetic diversity analysis of white maize inbred lines using SNP markers

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White maize (*Zea mays* L.) represents an important genetic resource with significant nutritional and agronomic value. Understanding its genetic diversity is essential for the conservation of local germplasm and the development of improved cultivars with desirable traits. A set of 63 white maize inbred lines sourced from the Gene Bank of the Maize Research Institute and its breeding programs was analyzed to assess genetic diversity and population structure using a 25K SNP Illumina Infinium Array (TraitGenetics). After quality filtering, 19,172 high-quality Single Nucleotide Polymorphism (SNP) markers were retained for analysis. Genetic diversity parameters were estimated using PLINK 1.9, while genetic distances and a dendrogram were generated in Tassel 5. Minor allele frequency (MAF) ranged from 0.05 to 0.5 (average 0.31), observed heterozygosity (H_o) from 0.015 to 0.1 (average 0.03), and expected heterozygosity (H_e) from 0.095 to 0.5 (average 0.40), indicating a relatively low level of diversity. Cluster analysis classified the inbreds into three main groups, with Group I comprising the majority of genotypes (39), Group II (19) and Group III containing only five. Principal Coordinate Analysis (PCoA) performed in R software supported these findings. Population structure analysis using STRUCTURE v2.3.4 confirmed the presence of three subpopulations (optimal $K = 3$), although some discrepancies in genotype grouping between cluster and STRUCTURE analyses were observed. These findings provide a comprehensive overview of the genetic relationships among the examined lines and highlight their potential as a valuable resource for future breeding programs and germplasm conservation strategies.

Keywords: white maize; genetic diversity; SNP array

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Variability of vitamin E content in ZP popcorn hybrids

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Vitamin E is a group of eight lipid-soluble antioxidants found in plants, divided into tocopherols and tocotrienols. As an essential nutrient, it supports seed development, stress response, and membrane protection. In humans, it helps protect cell membranes and may reduce the risk of diseases such as Alzheimer's, cancer, neurological disorders and inflammation. Both tocopherols and tocotrienols exist in four isoforms: α (alpha), β (beta), γ (gamma), and δ (delta), classified based on the position of methyl groups on the chromanol ring. This study analyzed α , $\gamma+\beta$, and δ tocopherols (T) and tocotrienols (T3) in five popcorn hybrids grown in Zemun Polje (ZP) and Kruševac (KŠ), Serbia. The content of vitamin E was higher in Zemun Polje, indicating that environmental factors had a significant impact on the content. The average content of α -T was 5.31 $\mu\text{g/g}$ dry weight (DW) at ZP and 4.27 $\mu\text{g/g}$ DW at KŠ, with ranges of 3.99–7.52 $\mu\text{g/g}$ DW and 3.18–5.03 $\mu\text{g/g}$ DW, respectively. The highest α -T content at both locations was recorded in hybrid ZP 6119k. The average content of $\gamma+\beta$ -T was 29.63 $\mu\text{g/g}$ DW at ZP and 23.51 $\mu\text{g/g}$ DW at KŠ. δ -T content ranged from 1.69–3.63 $\mu\text{g/g}$ DW at ZP and from 0.89–2.25 $\mu\text{g/g}$ DW at KŠ. The highest α -T3 content at both locations was found in hybrid ZP 614k (5.21 $\mu\text{g/g}$ DW at ZP and 3.62 $\mu\text{g/g}$ DW at KŠ). $\gamma+\beta$ -T3 varied from 4.13–7.07 $\mu\text{g/g}$ DW (ZP) and 3.29–6.99 $\mu\text{g/g}$ DW (KŠ). The highest values of δ -T3 were 0.74 $\mu\text{g/g}$ DW in ZP 6153k (ZP) and 0.76 $\mu\text{g/g}$ DW in ZP 6119k and ZP 6170k (KŠ). Popcorn hybrids with higher Vitamin E content, especially ZP 6119k, show potential as functional foods with added health benefits for human consumption.

Keywords: *specialty maize; tocopherols; tocotrienols*

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Genetic characterization of maize landraces using SSR markers: Insights for breeding applications

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This study aimed to evaluate the genetic diversity of traditional maize landraces and identify promising germplasm for breeding by performing molecular characterization of 31 maize landraces and 5 testers using Simple Sequence Repeat (SSR) markers. Of the 33 SSR primers initially tested, four were excluded due to monomorphism or low amplification efficiency, while the remaining 29 revealed a total of 139 alleles, with an average of 4.8 alleles per locus. The highest number of alleles (7) was observed with primers phi087 and phi080, whereas four primers (umc1265, umc1282, phi075, and umc1310) each detected the lowest number (3). Genetic distance analysis based on the Nei method revealed a high level of variability. Among the landraces, genetic distances ranged from 0.11 (AN2249 vs. AN2047) to 1.97 (AN1509 vs. AN1267). Population AN1509 exhibited the highest average genetic distance (1.643), suggesting strong divergence potential, while AN1945 had the lowest (0.478), indicating greater genetic similarity. For the five testers (L217, L73B013, L255/75-5, F2, and Polj17), genetic distances ranged from 0.43 between L217 and L73B013 to 1.54 between L217 and L255/75-5. Intermediate values were observed for F2 vs. L255/75-5 (0.64), Polj17 vs. L255/75-5 (0.57), and Polj17 vs. F2 (0.74), reflecting their differing genetic backgrounds and pedigrees. These results align with the known pedigree and origin of the testers. Principal Coordinate Analysis (PCoA) explained 35.9% of the total variation across the first two axes and revealed clear differentiation between the testers and most local populations. Population structure analysis (K=4) grouped the genotypes into four distinct clusters. The testers formed a homogeneous group, while the local populations exhibited greater intra-population diversity. These findings highlight the broad genetic base of the maize landraces and identify highly divergent genotypes. Among them, AN1509, with its pronounced genetic divergence, stands out as a valuable resource for future breeding programs.

Keywords: *maize landraces; genetic diversity; SSR markers; cluster analysis; breeding potential*

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Investigating the bioactive properties of microwave pretreated gluten hydrolysate fractions obtained via ultrafiltration

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A suspension of gluten from wheat was pretreated in a microwave reactor (Anton Paar Monowave 300) with the power of 200 W and afterwards subjected to enzymatic hydrolysis (final gluten concentration of 2% (w/w)) with Alcalase at 60 °C, while a control hydrolysis was performed without microwave pretreatment under the same conditions. The obtained hydrolysates were further separated by a pressurized ultrafiltration stirred cell (Millipore 8050 unit) with cellulose membranes with different pore sizes, 1–30 kDa. By consecutively applying several different cellulose membranes, we were able to perform a multistep separation process of hydrolyzed gluten peptides in order to investigate their bioactive properties, such as ABTS•+ and DPPH• neutralization, and Fe²⁺ chelating ability. The ABTS•+ and DPPH• neutralization properties of the produced gluten hydrolysates showed no significant differences among the tested fractions, while the ability to chelate Fe²⁺ ions is particularly noteworthy. Fe²⁺ ion chelating activities of the obtained fractions (F1-F5) showed that microwave pretreated hydrolysate fractions F2 (10-30 kDa), F3 (3-10 kDa) and F4 (1-3 kDa) were the main carriers of the bioactivity, with Fe²⁺ chelating activities ranging from 50–60%. A 10-25% increase of Fe²⁺ chelating activity was recorded for the microwave pretreated hydrolysate fractions in comparison to the control. It was concluded that the microwave pretreatment of gluten had significant effect on the Fe²⁺ chelating activity of the obtained hydrolysate fractions. No significant differences were recorded for the DPPH• neutralization, besides the control sample F1 (> 30 kDa) fraction. The ultrafiltration separation process showed that microwave pretreatment had improved the Fe²⁺ chelating activity, for the values recorded between the same fraction size of the two hydrolysates. The implementation of microwave pretreatment may be further utilized to produce effective smaller-sized bioactive peptides.

Keywords: *gluten; antioxidant activity; metal-ion chelating activity; bioactive peptides; enzymatic hydrolysis*

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Increasing EU project participation through stronger support structures: Insights from the SKUPP project

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The project “SKUPP – Cooperation to strengthen the effectiveness of project offices” aims to increase the capacity of administrative and research staff and to strengthen the role, position, and performance of Slovenian public research institutes in centralized EU programmes (e.g. Horizon Europe, COST). Research management is becoming increasingly demanding and requires the use of innovative working approaches, effective management of resources, and strategic thinking. The SKUPP consortium seeks to strengthen the support environment through effective project offices in public research organisations, improving their performance in applying for, implementing, and reporting on EU projects. The project is led by a consortium of four institutions: the Agricultural Institute of Slovenia, Geological Survey of Slovenia, Slovenian Forestry Institute, and Educational Research Institute. Since 2023, we have organised over 30 training courses, workshops, and networking events with more than 700 participants and hired 10 new project office staff in the consortium. Activities included training on writing proposals for ERC grants and Horizon Europe calls, financial management and auditing of projects, public speaking and communication of science, use of digital media and social networks, and the gender dimension in research. We have held several knowledge transfer workshops, workshops on the protection of intellectual property, and several meetings with National Contact Points (NCPs). To strengthen cooperation and shared learning, we hold annual consortium team-building events. The holistic approach of the SKUPP project, with its focus on research support and project management, is already showing promising results, as the number of proposals submitted to various centralised EU programmes has doubled in two years and ongoing projects are being managed more effectively.

Keywords: *research management; project office; capacity building; EU programmes; teamwork*

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Dissemination, communication and exploitation in the CREDIT Vibes project

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The CREDIT Vibes project aims to effectively communicate, disseminate, and exploit project results, implement strategic communication plans and support the exchange of knowledge, technology, and skills between partners and stakeholders. A comprehensive Communication, dissemination, and exploitation plan was developed to guide outreach and visibility efforts. This plan includes the creation of a unified visual identity, featuring the project logo, templates for various document types, and standardized text for press releases. Communication, dissemination, and exploitation plan will outline clear objectives, define the target audiences, craft key messages, and specify dissemination tools and indicators for evaluating the impact of these efforts. The strategy will ensure that various stakeholder groups are informed according to their specific interests and needs. Dissemination occurs continuously and extends beyond the project's duration through scientific publications and conference presentations. Communication activities include engagement via the Nutribreeding Hub, demonstration events, and social media (Twitter, LinkedIn, Facebook). All project partners actively contribute to dissemination and communication efforts at national, European, and international level by participating in scientific conferences, stakeholder events, and other targeted forums to maximize outreach and engagement. Knowledge management activities include the organization of conferences, workshops, and educational events. Notably, the International Maize Research Conference, organized by the Maize Research Institute, will feature a dedicated session on nutribreeding, showcasing project outcomes and expanding scientific collaboration. In Addition, demonstration events such as Fields of Innovation and Open Labs are aimed at local farmers, breeders, advisors, and seed companies, helping translate research into practice.

Keywords: *knowledge exchange; communication activities; EU programmes*

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The Greek Living Earth: A National Digital Infrastructure for Soil Carbon Monitoring and Climate-Resilient Agriculture

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The urgent need to mitigate climate change and support sustainable land management requires innovative, data-driven approaches to monitor and enhance soil health. The Greek Living Earth (GLE) addresses this challenge by developing a national digital infrastructure for the integration, storage, and fusion of geospatial hyperspectral and field data, for monitoring of soil carbon stocks and fluxes across Greece. At its core, *the GLE* will establish a digital soil twin which will be achieved by fusing heterogeneous data sources—satellite hyperspectral and very high-resolution multispectral imagery, in situ measurements from IoT sensor networks, UAV imagery, and diagnostic field toolkits. Using AI and interpretable ML models, these datasets will be harmonized within an open-source data cube infrastructure, producing thematic maps of indicators. It will directly support carbon farming and sustainable agriculture by providing farmers, policymakers, and other stakeholders with robust, actionable tools. Producers will be able to assess soil health, adopt carbon sequestration practices, and certify climate-smart farming methods through reliable and traceable data services, enabling Greece to meet its EU obligations under the Common Agricultural Policy and also open new opportunities for farmers to access voluntary carbon markets through certified carbon credits. By bringing together expertise in Earth observation, soil science, artificial intelligence, and digital infrastructure, *The GLE* aims to reach a high level of technological readiness (TRL 8), transitioning from research to market-ready services. Beyond its scientific and technological innovation, the project aspires to foster a paradigm shift in Greek agriculture: from input-intensive practices towards resilient, carbon-positive systems contributing to climate mitigation while sustaining rural livelihoods. Ultimately, *The GLE* acts as a living laboratory for the digital transformation of environmental monitoring and carbon farming in Greece, demonstrating how interdisciplinary collaboration and cutting-edge digital technologies can be mobilized to address pressing environmental challenges, strengthen the bioeconomy, and contribute to climate goals.

Keywords: digital soil twin; carbon farming; Earth observatio; AI; sustainable agriculture

Land suitability classification for agricultural production in Slovenia based on natural attributes

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The processes of regionalisation and zoning of agricultural land involve the systematic classification and evaluation of the land suitability for different types of agricultural production based on an analysis of natural conditions and economic parameters. These processes aim to optimise land use, promote sustainable resource management and support the adaptation of agricultural practices to climate change. The RajonSI project, launched in autumn 2024 and scheduled to run for three years, aims to enhance agricultural productivity and provide information for sustainable spatial planning in Slovenia by assessing land suitability according to production type. The project integrates different data sets, including soil properties (pH, organic matter content, texture, bulk density, stoniness), climate variables (precipitation, temperature, evapotranspiration), topographic features (elevation, slope, aspect, topographic wetness index) and additional environmental and spatial data (flood-prone areas, water protection zones, land use). The climate variables are modelled for several scenarios, which enables the assessment of land suitability for the future. The suitability assessments are tailored to general land use categories – arable land, orchards, vineyards and permanent grassland – as well as major crops such as maize, cereals (wheat and barley), potatoes, apples and pears, strawberries, blueberries and pastures. Using GIS-based methods and spatial algorithms, the project will produce detailed maps of agricultural suitability for Slovenia. The overall aim of the project is to identify critical factors that influence successful agricultural production and to develop spatial information layers that support an evidence-based decision-making tool for stakeholders, including the Ministry of Agriculture, Forestry and Food and local farmers. This will also take into account the lessons learnt from the HIGHLANDS.3 project, which focused on a collective approach to research and innovation for sustainable development in the highlands. The RajonSI contributes to the sustainable use of natural resources and provides a basis for increasing agricultural productivity under changing environmental conditions.

Keywords: *land suitability assessment; agricultural zoning; geospatial analysis; climate change adaptation; sustainable agriculture*

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Remote sensing and AI-based monitoring of soil properties for Tier-3 MRV systems in mediterranean agroecosystems

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This study presents a robust methodology for spatially explicit monitoring of key soil properties in Mediterranean agroforestry and cropland systems using remote sensing (RS) and artificial intelligence (AI). Developed within the MRV4SOC project, the approach supports Tier-3 Monitoring, Reporting, and Verification (MRV) by producing high-resolution geospatial layers of topsoil characteristics critical for modeling Soil Organic Carbon (SOC) dynamics. The framework was applied at two long-term demonstration sites in Spain: *Quercus ilex* Dehesas in Extremadura and rainfed cereal systems at La Canaleja experimental station. Multi-temporal Sentinel-2 data (2018–2022) were combined with machine learning models to predict SOC, pH, sand, silt, clay, and Total N. Ground-truth data were primarily sourced from LUCAS 2018 due to limited site-specific sampling, and a composite of bare soil reflectance from Sentinel-2 bands (B02–B12) at 20 m resolution was used for geospatial prediction. Quantile Regression Forests (QRF) and XGBoost were applied, with QRF performing best in agroforestry systems. Model evaluation used R^2 , RMSE, nRMSE, and RPIQ. Excellent predictive performance was achieved for pH ($R^2 = 0.76$, RMSE = 0.044, RPIQ = 3.15), good accuracy for clay ($R^2 = 0.60$, RMSE = 5.56, RPIQ = 2.41) and Total N ($R^2 = 0.54$, RMSE = 0.045, RPIQ = 2.21), and fair but useful results for sand ($R^2 = 0.51$, RMSE = 9.72, RPIQ = 1.82) and silt ($R^2 = 0.66$, RMSE = 8.60, RPIQ = 1.78). SOC prediction showed moderate variance explanation ($R^2 = 0.63$) but higher relative error (RMSE = 1.06, nRMSE = 109%, RPIQ = 1.48), reflecting challenges in estimating low SOC levels. XGBoost performed better for some properties in croplands and for nitrogen and texture variables in agroforestry. Prediction uncertainty was assessed using the Prediction Interval Ratio (PIR) at landscape and sub-landscape levels. While moderate to low for most variables, higher variability occurred for sand and silt in croplands and SOC in agroforestry. These results demonstrate the potential of integrating EO and AI for cost-effective, high-quality soil monitoring and highlight the need for increased ground-truth data to enhance accuracy and reduce uncertainty.

Keywords: remote sensing; soil properties; AI modelling; SOC monitoring

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