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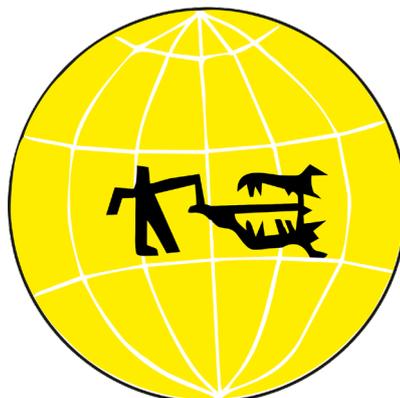
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Soil ecosystem services in a climate change context

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Summary

Soils are the base of life and essential for well-being. Since historical times they have played an important role in human evolution and provided the basis for human sedentarism. Most first civilizations were developed in flood plains (Tigre and Euphrates rivers- Mesopotamia; Indus and Ganges-India; Yellow River-China) where soils are fertile. Therefore, it is impossible to dissociate our evolution from the soil's role. However, due to intensive exploitation and climate change, several civilizations collapsed (e.g., Mesopotamia). This calls attention to the effects of unsustainable practices coupled with climate change on the ecosystem services soil can supply. It is well known that farming practices can trigger soil degradation and hamper soil productivity. Nevertheless, is essential also to understand how climate change can trigger these effects. The increased extreme climate events (droughts and floods) will decrease soil functions, ultimately affecting the services supplied to the soils. For instance, combining dry periods and intensive precipitation will decrease the soil's capacity to regulate erosion and floods or purify water. This will also negatively impact carbon sequestration and the soil's capacity to contribute to microclimate and air quality regulation and pollination. Degraded soil will not sustain a healthy environment and will contribute to the degradation of other ecosystem services, such as recreation or landscape aesthetics. Climate change and extreme events' increasing frequency and intensity will increase soil degradation. The key is how we can minimize this. Many challenges are ahead such as halting and restoring agricultural ecosystems or establishing more sustainable practices (e.g., nature-based solutions). In this talk, I will overview the problems associated with climate change's impact on soil ecosystem services and how we can mitigate their effects.

Keywords: Soils, ecosystem services, climate change, extreme events, degradation

Leveraging insect genetics and breeding for sustainable protein production in the 21st century

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Summary

Agriculture is under increasing pressure from two sides. On one side, we need to produce more food for the growing human population, which is at the same time shifting towards a protein-rich diet. On the other side, climate change is reducing the success of production with increasing weather volatility. One way to contribute to sustainable protein production is to incorporate insects as one of the key pieces of the larger jigsaw that is agriculture and our food system. Insects are a part of the human diet in many parts of the world. There are about 2 billion people that eat insects or insect-based products regularly, often for their high protein content. However, this insect production has a large untapped potential from at least five perspectives. First, insects generally have lower requirements than livestock regarding feed, water, and space. These lower requirements mean that there is potential to produce protein in more sustainable ways. Second, while there is a lack or complete absence of eating insects in many communities, insects can be used more than just as a source of protein-rich food for people; they can also be used as a source of protein-rich feed for livestock and fish. Third, when reared for feed, insects can valorise vast organic by-product and waste streams in the food system and promote local bioeconomy. This waste valorisation has two important knock-on effects on sustainability. We waste almost a third of grown food before reaching our households. Further, by valorising these waste streams, we could reduce the pressure on deforestation to get new arable land (say, for soy production) and on overfishing to get sufficient protein feed for livestock and fish. Fourth, many insect species could be farmed at a large scale for different purposes (food, feed, feedstocks for biotechnology, etc.) in many parts of the world. Fifth, there is also ample genetic diversity within insect species, so we could selectively breed improved populations that could further boost sustainable protein production.

Black soldier fly

Within this rapidly growing new sector of insect farming, the black soldier fly is emerging as a leading species for the insect as feed sector. The black soldier fly, *Hermetia illucens*, could replace unsustainable fishmeal and soymeal in aquaculture and livestock feeds, reducing overfishing and deforestation. For example, the UK imports almost 100K tonnes of fishmeal and 2.2M tonnes of soymeal per year and feed accounts for ~70% of pig and poultry production costs. Insect production could contribute to the UK net-zero targets and ensure that the UK agri-food supply chain is not disrupted by global market volatility and climate change. However, insects as feed cannot yet compete on price-point with fishmeal or soymeal. Two developments that will make insects as feed competitive are improved production, by scaling, and improved genetics, by selective breeding. Scaling is underway - commercial facilities producing up to 15K tonnes of insect protein and 5.3K tonnes of insect fat per year are in operation. In this presentation, I will focus on leveraging breeding and genetics to boost sustainable protein production.

Black soldier fly breeding and genetics

The black soldier fly has been only recently domesticated for farming but is already becoming a valuable protein producer. Looking at the history of improvement in other agricultural species, with gains of 10-100x compared to wild ancestors, I envision an extraordinary future for black soldier fly breeding. Black soldier fly originates from the Neotropical ecozone of America but has spread to all continents via overseas trade. Black soldier fly has similar properties as species in aquaculture and plant breeding. In

these settings breeding programmes are based on population improvement and product development. Population improvement is driven by defining breeding targets (larval and fly traits), data recording, intense and accurate selection, management of genetic diversity and fast breeding. The black soldier fly has immense potential for population improvement due to its large fecundity (up to 1,000 eggs per female), high genetic diversity (overall $F_{st}=0.24$ and max-pairwise $F_{st}=0.6[5]$), and short generation interval (6-7 weeks under ideal conditions). Practical challenges lie in managing numerous fast and short-living individuals and recording and leveraging data to drive accurate breeding decisions. These challenges will be addressed with specialised breeding technology and know-how, high-throughput phenotyping at scale using automation, vision, possibly genome-wide genotyping, and data science.

Product development is driven by defining production targets and assembling genotypes that deliver these targets. In black soldier fly, I envision crossbreeding that will leverage the wealth of black soldier strains to target a range of production settings in the Global North and the Global South. Population improvement of the strains into high-performance breeds will drive continuous product development and intellectual property protection.

There are also three major risks that breeders will have to manage. First, intense selection in prolific species can deplete genetic diversity and lead to inbreeding depression. Second, diseases of black soldier fly are largely unknown. Third, the microbiome plays a key role in some phenotypes, suggesting that breeders will have to account for these interactions while improving their populations.

In the presentation, I will highlight some of the recent results from a research collaboration with a commercial black soldier fly breeding programme, including challenges and perspectives on how to address these challenges.



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Modeling the cost-effectiveness of direct replacement of mineral forms of N, P and K with organic fertilizers

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Summary

The cultivation of field crops in recent decades has been marked by the negative impacts of climate change, increased risk of soil degradation and the risk of environmental stress, and the latest challenge is the increase in the price of energy sources and mineral fertilizers. At the same time, in the Republic of Croatia there is a large proportion of arable soils with low SOM content, like in the soils analyzed in 2020, almost half of the soil was found to have less than 2% SOM with an average content of <1.6%. On the other hand, the development of sustainable agriculture and digital technologies enables not only the precise application of fertilizers, but also the precise modeling and calculation of the optimal fertilization and the profitability of fertilization including mineral and/or organic fertilizer. Therefore, we developed a model to help make decisions in fertilization based on the cost-effectiveness of replacement of mineral fertilizers (containing N, P and K) with organic fertilizers. The model does not consider the cumulative fertilizing and improving effect of organic fertilizers on soil fertility, but only the direct value of N, P and K in the organic fertilizer. The model is based on data on organic fertilizer (N, P, K content and dynamics of N availability), data on soil fertility and crop needs (for calculating optimal amounts of N, P and K in fertilization), data on plot distance and available mechanization (for calculating the costs of transport and application of organic fertilizer) and the prices of mineral and organic fertilizers (for total costs of mineral and organo-mineral fertilization). The result of the model is the calculation of the total costs of fertilizing only with mineral fertilizers and with a combination of mineral and organic fertilizers, and an analysis of the cost structure (fertilizer purchase cost, transport cost, and fertilizer application cost) depending on the price of the fertilizer (mineral and organic), nutrient content (N, P and K), distance and the size of the arable plot, the method of transport, the capacity of the spreader, the cost of the employee's work, etc. The model allows comparison of profitability by simply changing the value of one, several or all input variables. The result contains information on the share of transport and application costs in total fertilization costs, for example a share of 23.7% if 0.2% N in fertilizer and plot distance of 0.5 km and a share of 8.5% if 0.8% N in fertilizer. Also, the models calculate the inflection points when organo-mineral fertilization becomes cheaper than mineral fertilization. The inflection point can be expressed as minimum nutrient content in manure (N, P and/or K), as maximum plot distance, minimum spreader capacity or as a combination of different values. For example, in fertilizing corn on medium-fertile soils inflection point is when organic fertilizer contains 0.5% N, and in fertilizing wheat it should be at least 0.64% N with the appropriate proportional content of P and K.

Keywords: manures, transport costs, manure application, nutrient content, inflection point

Spatio-temporal change of soil pH, P₂O₅ and K₂O content under variable mineral nitrogen fertilization

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Summary

Soil sampling is the first step in generating field-specific information on which to base agricultural input decisions. The objective of the study was to evaluate spatial and temporal variability of soil pH, plant available phosphorus (P₂O₅) and potassium (K₂O) content under variable mineral nitrogen (N) fertilization. The research was conducted on the experimental field within hydro-ameliorated cropland in the Western Pannonia subregion of Croatia (45°33'N, 16°31'E). Grid soil sampling (15x15 m; n=200) was carried out with semi-automatic circular tractor soil probe at 30 cm depth after winter wheat harvest in 2010 and 2016 covering ten fertilization treatments (0-300 kg N ha⁻¹). A sequence of spatial changes in soil properties was influenced by differences in N rates, liming, field drainage and specific terrain features, although similar spatial response was apparent for both investigated years. A two-way ANOVA revealed that only soil pH values significantly differed among two years (p<0.05), while significant effect of treatment was recorded for soil P₂O₅ and K₂O content. Soil pH and P₂O₅ values were higher in soils sampled in 2016 in addition to samples collected in 2010 due to the liming applied in mid-term (max. of 21% and 24%, respectively). In 2016, soil K₂O content was lower on well fertilized treatments compared to 2010, which can be attributed to high winter wheat yield in 2016. Interaction time x N treatment was not significant for investigated soil properties. Precision sampling revealed complex relations in soil properties presumably conditioned by more agroecological factors.

Keywords: arable farming, mineral fertilization, precision soil sampling, soil quality, Stagnosols

Optimization of soil management by modeling the availability of iron and zinc

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Summary

The development of digital technologies enables a more thorough use of available data for optimizing crop production. Thus, results of soil analyzes can be used to create models for estimating soil properties that have not been analyzed. An example is a model for prediction the availability of Fe and Zn based on basic soil properties ($\text{pH}_{\text{H}_2\text{O}}$, pH_{KCl} , SOM). However, it is necessary to determine the accuracy of such models by validation. The aim of this research was to validate models for predicting the availability of soil Fe and Zn. Previously created models based on the analysis of 229 soil samples were chosen for validation, a model based on 3 basic soil properties was used to estimate Fe availability, and a model for Zn availability that additionally includes the soil total Zn content ($\text{Zn}_{\text{EDTA}} = 0.0634 \times \text{Zn}_{\text{AR}} - 1.012 \times \text{pH}_{\text{H}_2\text{O}} + 0.807 \times \text{pH}_{\text{KCl}} - 0.095 \times \text{SOM}$). For validation purposes, the basic properties ($\text{pH}_{\text{H}_2\text{O}}$, pH_{KCl} and SOM) of 40 soil samples were analyzed with additional analysis of the available (extracted by EDTA) and the total Fe and Zn content (extracted by aqua regia - AR). In the validation set, the content of available Fe was in the range 9.4-150.2 mg/kg and available Zn 1.9-12.5 mg/kg. Thus, laboratory analysis of validation set determined 57.5% of soils with low availability and 12.5% with high Fe availability, while there were no soils with low Zn availability and 60% of soils were with high Zn availability. However, it was determined that the validated models were not precise enough to predict the availability of Fe and Zn, because the models overestimate the availability of Fe (high availability of Fe was estimated in 97.5% samples) and underestimate the availability of Zn (low availability of Zn was estimated in 55% samples). For this reason, new models were tested that were more accurate. The most accurate model for Fe was model using additional data on the content of total Fe and soil texture ($\text{Fe}_{\text{EDTA}} = 0.0043 \times \text{Fe}_{\text{AR}} - 12.525 \times \text{pH}_{\text{H}_2\text{O}} + 9.968 \times \text{pH}_{\text{KCl}} - 6.155 \times \text{SOM} - 1.246 \times \text{Texture}$) estimating 60% of soils with low Fe availability (should be 57.5%) and 40% of soils with high Fe availability. On the other hand, all new models for predicting Zn availability had a significant error (on average about 41 %) due to the estimation of a very low range of Zn availability, 2.2 - 5.7 instead of 1.9-12.5 mg/kg (the best model was $\text{Zn}_{\text{EDTA}} = 0.0334 \times \text{Zn}_{\text{AR}} + 0.028 \times \text{pH}_{\text{H}_2\text{O}} - 0.056 \times \text{pH}_{\text{KCl}} + 0.418 \times \text{SOM} + 0.507 \times \text{Texture}$). We can conclude that in order to create a model for estimating the availability of Fe and Zn, it is necessary to include a significantly larger initial set of samples with a wide range of values of basic soil properties and availability of Fe and Zn.

Keywords: modeling, soil Fe availability, soil Zn availability, aqua regia, EDTA extraction

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Inputs in organic agriculture – new in EU legislation and examples of area regulation in selected countries

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Summary

Organic agriculture (OA) is a sustainable form of agriculture and is an example / model of a circular economy. Clear rules are uniformly defined in the European Union (EU) in legislation 2018/848. The OA is the most limited of all production systems in terms of the use of extra-farm inputs which are listed in regulation 2021/1165. The OA is a system that contributes to the integration of environmental protection requirements into the common agricultural policy and promotes sustainable agricultural production, as well as directly contributing to the achievement of the European Union's environmental policy objectives. It has gained special importance based on the "From Field to Fork" strategy, which sets the goal of achieving at least 25% of agricultural arable land under OA by 2030. Knowledge and availability of suitable inputs for the OA can enable the development of more demanding agricultural branches in organic production and animal husbandry as well as processing, where it is necessary to choose e.g. also suitable cleaners, packaging... Within the national research project "Inputs in organic agriculture", several analyses of the situation in the field of management and information on variable inputs for organic farming abroad and the situation in Slovenia is reviewed including some of the results of ongoing projects recently held on this topic in EU. Based on previous experience, analysis of methods of work and organization in selected countries, interest of providers / distributors, in cooperation with the competent expert's proposal / concept for establishing a system - a model for assessing inputs and their suitability for OA is discussed. The availability of information on inputs for organic farming could facilitate the decision to switch new farms from conventional to organic farming, thus contributing in practice to the achievement of national and EU OA development goals. Using verified inputs, growers, processors, control organizations and consumers will have the assurance that the principles and rules of organic farming are followed in practice. The aim of the contribution is to protect the integrity of organic farming and that the principles of organic farming are observed even when using selected inputs for OA among operators.

Keywords: organic agriculture, inputs, legislation

Testing varieties for organic production - some cases from vegetable and field crops production

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Summary

Organic production is dominated by varieties that have been bred for conventional production. Experience shows that some conventional varieties can also be used in organic production, but most of these varieties do not have enough emphasis on traits that are very important for organic production such as competition against weeds, resistance to damage caused by mechanical means of weed control, better nutrient utilization efficiency, resistance to seed-borne diseases, and resistance to diseases and pests. Also organic varieties, which are bred with aims of organic farming, are usually tested as a part of a regular value for cultivation and use (VCU) testing or special variety testing within public services on conventional fields. There are only a few countries where for some species in varieties testing organic farming is used as a production method. The aim of this contribution (and of the national research project CRP V4-2007) is presenting a part of preliminary results of evaluating protocols of VCU testing of different crops in field experiments under organic farming principles. In 2022 on two sites (Kranj, Maribor) were tested varieties of Batavia lettuce (10 varieties of organic seeds and 5 varieties using non-treated conventional seeds), 8 varieties of potato in Jablje, 11 varieties of winter wheat and one spelt variety in Jablje, 5 varieties of maize and 5 varieties of soybeans were tested in Jablje and in Maribor. Based on preliminary results and evaluation of testing protocols some recommendation have been prepared for improvement agricultural techniques on the experimental fields and for improving testing protocols for different crops according to the organic production method.

Keywords: organic production method, testing varieties, protocols

Tillage, biochar and farmyard manure impact soil physical properties and maize yields on Cambisols in Croatia

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Summary

The negative environmental impact of conventional agriculture threatens agroecosystem stability and food security. In this context, searching for optimal soil management is crucial for maintaining and improving soil functions. This work aims to determine the impact of conventional tillage (CT) and reduced tillage (RT) on Cambisols in a semi-humid environment in Marija Magdalena (Croatia). Under each tillage treatment, subplots were biochar – B (40 t ha⁻¹), farmyard manure – FYM (40 t ha⁻¹) and control - C. The impact was assessed on physical soil properties and maize (*Zea mays*) yield. Soil bulk density (BD) at 0-15 cm depth was significantly lower in RT-B when compared to the CT-B. At 15 – 30 cm depth, BD was significantly lower in all RT sub-treatments than in CT. Penetration resistance (PR) did not exceed 2 MPa. At 0-15 cm, PR was significantly lower in RT-C than in CT-C and CT-FYM plots. Between 15-30 cm, PR was significantly higher in CT-B and CT-C than in the other treatments. Soil water content (SWC) was high in RT treatments at 0-15 cm. At 15-30 cm, SWC was significantly lower in CT-B than in RT-B. Water-stable aggregates were higher in B and FMY plots under RT than under B and FMY plots under CT. Moreover, maize grain yield was higher in RT treatments than in CT. Specifically, FYM addition with CT showed a better impact than FMY with RT. Maize respond oppositely when compared biochar and tillage treatments, RT-B was better management than CT-B. Regarding short-term results, we can conclude that RT is an advisable alternative for CT due to its positive impact on soil physical condition. Efficient management for stable yields and amendment recommendations could be determined by implementing this research over an extended period.

Keywords: reduced tillage, soil amendments, soil compaction, grain yields

Conventional and conservation soil management impact on soil properties, runoff generation and element losses in horticultural cropland in Dalmatia (Croatia)

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Summary

This research aimed to determine the effect of different soil management in horticultural cropland on a slope. The study is conducted in Polača (44°00' N; 15°29' E), Croatia, on clay with rock fragments soil Calcaric Leptosol. Rainfall simulation experiments (intensity 60 mm h⁻¹), measurements and soil sampling (0-10 cm) were conducted in three seasons (Spring, Summer, and Autumn) on 4 studied treatments. Tilled, barley straw mulched (2.5 t ha⁻¹), hay mulched (2.5 t ha⁻¹), and grass cover to determine soil physical and chemical properties, hydrological response, and pollutant losses. The bulk density (BD) was not significantly different between the treatments in all three seasons, while seasonally, the re-compaction status increased from Spring (1.4 g cm⁻³) and Summer (1.5 g cm⁻³) to Autumn (1.7 g cm⁻³). Water stable aggregates (WSA) and soil organic matter (SOM) were significantly higher on the grass-covered and mulched treatments than at tilled treatments. Significantly higher copper (472 kg ha⁻¹), potassium (16045 kg ha⁻¹), and phosphorus (1413 kg ha⁻¹) concentrations in soil were noted on the tilled treatment in Spring but lower in Summer and Autumn when compared to grass-covered and mulched treatments. Time to ponding (TP) and Time to runoff (TR) were significantly higher on the Hay treatment in Spring (330 sec, 1800 sec) and Autumn (193 sec, 672 sec), while in Summer, there were no differences between the treatments. This affected the soil loss (SL), which was significantly higher on the tilled treatment in Spring (497.07 kg ha⁻¹) and Autumn (176.31 kg ha⁻¹). Results obtained in this study show in Autumn that the application of barley straw, hay and permanent grass cover had increased SOM, WSA, TP and TR, while it reduced P losses (by 269.11 %), K losses (by 153.78 %) and Cu losses (by 115.82 %) compared to tilled treatment. The results showed that barley straw, hay and permanent grass coverage are effective nature-based solutions to prevent soil loss.

Keywords: soil management, trace elements, rainfall simulator, mulching

Impact of environmental conditions on the growth, development and certain chemical properties of wild garlic (*Allium ursinum* L.)

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Summary

It is known that wild garlic has been used in traditional medicine since ancient times, but studies on its composition and pharmacological effectiveness are recent and very rare. Young leaves, stems and bulbs are used as a salad or spice, and contain essential oil, vitamin C, alliin and fructan. In traditional medicines, it is used for the treatment of skin diseases, stomach and intestinal problems, diseases of the respiratory system and arteriosclerosis. For eating, the leaves are harvested before flowering, with a note that the tastiest are the very young leaves, before the flower stalk emerges. In addition to being used in cooking, this plant has medicinal properties and the mineral composition depends on the content of biogenic elements in the soil. Wild garlic often grows in groups forming a dense cover in beech, oak and hornbeam forests on soils rich in organic matter, with a moderately acidic to basic reaction. The aim of the work was to determine the influence of some soil chemical characteristics of a certain micro location on the morphometric properties of the wild garlic and the mineral content in the leaf in the vegetative and generative growth phases. During the wild garlic growing season in 2022, morphometric measurements were performed on 20 plants at five different micro locations (L1, L2, L3, L4, L5) in the area of the town of Sveti Ivan Zelina, Croatia. An average soil sample was taken from each micro location and leaf samples were taken for chemical analysis on two periods (before flowering and during flowering). At the investigated locations the soils had an acidic to weakly acidic reaction, with a good to very rich supply of humus. The supply of plant-available phosphorus in soil samples was poor, while for potassium the supply was good to rich. Significantly smaller leaf area before flowering was found at site L2 compared to other sites, while significantly larger leaf area during flowering was recorded at site L5 compared to site L1. A significantly higher content of vitamin C was determined in the leaves of plants before flowering (56.17 mg/100 g of plant material) compared to the content of vitamin C in the leaves of plants in bloom (27.88 mg/100 g of plant material).

Keywords: wild garlic (*Allium ursinum* L.), soil, morphometric measurements, vitamin C

Leaf apparatus as stress indicator in *Lactuca sativa*

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Summary

With the goal to compare the measurement of qualitative and quantitative parameters to the measurement of the fluorescence of leaves by OS 30ps and the level of SPAD index, research was conducted on lettuce. A random block design was used to set up the experiment with three replications and three distinct treatments. The first treatment entailed spraying an aqueous suspension of Aloex Agro stimulants on seedling roots 24 hours before transplantation, while the second treatment involved performing the same procedure with Sinergon 3000 instead. The treatments that were used were contrasted with the stimulant-free control group. Measurements made throughout the investigation included above- and below-ground plant mass, dry matter content of above- and below-ground plant sections, phosphate and potassium content, chlorophyll a, b, and carotenoid content, SPAD index, and fluorescence parameters Fm, Ft, Fo, and Fv / m. Aloex Agro was used to produce the maximum yield of 247.7 g of plant-1. Other qualitative and quantitative parameters had values that were noticeably greater than those found in the control treatment, indicating that these bio stimulators have greatly improved plant metabolic activity, which is reflected in the measured values. One of the simplest ways to get information on the state of crops in the field is undoubtedly through the use of these devices for plant analysis and diagnostics, but their application in this field has some limitations. The use of these tools for plant analysis and diagnostics is undoubtedly one of the straightforward ways to gather information about the health of crops in the field, but more research is required to establish their accuracy and variability in relation to variety, environmental factors, measurement, and interpretation of optimal ranks.

Keywords: *Lactuca Sativa* L., yield, fluorescence, SPAD index, bio stimulators

Bioavailability of soil Selenium in Southeast Europe can be modelled by soil properties

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Summary

Our soil survey of Southeast European (SEE) soils showed that the concentrations of selenium (Se), an essential human and animal micronutrient, are well below the deficiency level of 0.5 mg/kg. As Se uptake is mostly governed by plant food and feed intake, another valuable consideration is the Se bioavailability. The aim of this work was to analyze the soil chemical properties affecting Se bioavailability through the linear modelling procedure. The 52 samples of soil from arable soil layer (0-30 cm) were collected from agricultural fields in Croatia (Osijek), Serbia (Novi Sad) and Bosnia and Herzegovina (Sarajevo, Mostar, Banja Luka and Prud). Sampling was carried out using a diagonal sampling method in 20 to 25 stabs with footstep soil probe. Soil samples were dried and subject to chemical analysis. Selenium water solubility was used as a proxy of its bioavailability. We carried out partial least squares (PLS) regression analysis of Se_{H_2O} showing high predictive accuracy of validated models ($R^2=0.776-0.902$) depending on predictors, with highest loadings observed for loss of ignition (LOI), cation exchange capacity (CEC), total C and total N, along with expectedly high loading value for Se_{Tot} . Our results suggest that along to soil organic amendments improving the favorable soil properties, biofortification of cereal crops in SEE would be a worthwhile endeavor with possible broad socioeconomic and health implications.

Keywords: Selenium, biofortification, modelling, Southeast Europe

Agrochemical indicators of soil fertility in the Republic of Croatia in 2021

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Summary

The Croatian Agency for Agriculture and Food, in cooperation with authorized agrochemical laboratories, systematically organizes and conducts soil analysis in the Republic of Croatia. In total, 37,453 samples were analyzed over the past 3 years, but in 2021 almost three times more samples (27,716) were analyzed than in 2020 (9,869) and 2019 (9,868). However, the average results were very similar in all three years, since about 50% of the soils were either poor in humus (SOM) or poor in available phosphorus or soil acidity was increased. Somewhat better indicators in 2021 were determined only for the SOM content, even though as many as 41.8% of soils have <2% SOM (55.4% in 2019 and 48.8% in 2020), but the average SOM in soils with <2% SOM was almost identical to previous years (1.61%). That, unfortunately, means that in 2021 a low SOM content was determined in more than twice the number of samples (11,438) than in two previous years (4,817 and 5,467). Significantly more acidic soils (49.9% acidic or very acidic soils) were analyzed in 2021 than in 2020 (46.0%) or 2019 (44.5%) with less alkaline (21.1%) and neutral soils (13.3%) than in previous years. It is very important to emphasize that a significantly higher proportion (50.7%) of phosphorus-poor soils (classes A and B) than in previous years (44.8% and 37.3%) were determined, implying less moderate (22.5% vs. 26.7% - 30.1%) and richly P-supplied soils (26.8% vs. 28.5% - 32.6%) in 2021. The same trend, although of a slightly weaker intensity, was also noticed for classes of K availability in the soil, since an increase in the share of poorly supplied soils (29.1% vs. 23.3-19.3%) with a decrease in the share of moderate (48.4% vs. 53.7-55.4%) and richly supplied soils (22.5% vs. 23.1-25.4%) were determined in 2021. The results of the analysis indicate that the biggest threats to soil fertility, as in previous years, are insufficient SOM content, soil acidity and insufficient available phosphorus.

Considering that 21.4% of the soils were simultaneously poor in SOM and phosphorus, 19.7% of the soils were acidic ($\text{pH}_{\text{KCl}} < 5$) and poor in SOM, of which 11.4% were the soils of the lowest fertility due to the low SOM, acidity and the lowest class of available phosphorus, the most important for soil fertility, as in last year, are organic fertilization, optimization of P fertilization and liming of acidic soils.

Keywords: humus, soil pH, phosphorus, potassium, acidity

The influence of potassium fertilization on the properties of cereal straw for reinforcement of earthen architecture

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Summary

Cereals are the most important field crops with aspect of harvested area and production in the world. Grains are used as food for human and livestock, as well as raw material for many processing industries. However, cereal straw (stem) can also be used as reinforcement for earthen architecture (e.g. structures made of rammed earth, adobe or cob). The main goal of this study was to determine the influence of different potassium fertilization on the morphological properties (height and diameter of stem, nodes number, stem mass, and ear mass) but also chemical composition (K, N, P, Ca) of wheat, barley, oat and ray. Field experiment was set up during 2021/2022 vegetation season in three different doses: control, 60 kg ha⁻¹ K₂O and 120 kg ha⁻¹ K₂O. Monthly amounts of precipitation and mean air temperatures were monitored. Compared to the control treatment, an increase in morphological properties was observed for all cereals to which the fertilization was applied, as follows: stem height by 5.8 %, diameter at the stem base by 6.8 %, diameter in the middle of the stem by 7.7 %, nodes number by 2.4 %, stem mass by 63.5 %, and ear mass by 6.0 %. Furthermore, the following increase was also observed in: K and Ca by 9.4 %, P by 4.1 %, and N by 6.2 %. In general, cereals responded positively to fertilization, with an increase in the morphological values and chemical composition with respect to all investigated properties, except carbon. This can be important for usage of straw as reinforcement in rammed earth structures, which is being investigated within ongoing tests on rammed earth samples reinforced with the natural fibers.

Keywords: cereal straw, morphological properties, chemical composition, potassium fertilization, natural reinforcement, rammed earth

Acknowledgement: This study was a part of HRZZ project (RE-forMS - UIP-2020-02-7363): “Rammed earth for modelling and standardization in seismically active areas”, project manager: Assoc. Prof. Ivan Kraus (Josip Juraj Strossmayer University of Osijek, Faculty of Civil Engineering and Architecture, Osijek).

Value creation and ecosystem services of European Seaweed industry by reducing and handling potentially toxic elements (PTE) from breeding to soil

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Summary

The new BlueBio project - SeaSoil will deal with pressing issues regarding utilization of the blue biomass, seaweed, to promote a sustainable and competitive blue bioeconomy in Europe. The low trophic seaweed may significantly contribute to the food system as well as the carbon sequestration and storage (CSS) in agricultural soil when potentially toxic elements (PTE) of e.g. inorganic arsenic (As), cadmium (Cd) and iodine (I) can be managed safely in the food supply system.

The project is a consortium of ten European partners from Norway, Ireland, Denmark, Estonia and Croatia. The project goals will be reached through six work packages. The project goals are: a) to estimate heritabilities and genetic correlations in contents of Cd, As and I in sugar kelp, b) to study the impact of seaweed application rate and water saturation on the As dynamics in the soil to determine chemical reactivity, and potential bioavailability of labile As, c) to estimate the potential of seaweed amendments for CSS in agricultural soils, d) to study the impact of seaweed production on the environment using LCA, and conducting a cost-benefit analysis of the seaweed industry, e) to study the economic feasibility, and regulatory incentives, for production and use of (residual) biomass from farmed seaweed and f) to ensure the multi-actor approach and integrated cooperation, communication and human capacity building in line within Responsible Research and Innovation (RRI).

The overall goal is to investigate the potential of using seaweed in agriculture where the Faculty of Agrobiotechnical Sciences has the task of studying soil respiration of soil amended with seaweed as well chemical and physical soil properties of such amended soil.

Keywords: BlueBio, Seaweed, agriculture, carbon sequestration, PTE

Evaluation of enzymatic activity in soil after application of innovative fertilizer mixtures

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Summary

Due to the fact that the use of various types of fertilization changes the chemical, physical and biological properties of the soil, the need arises to study and control the effect of the applied fertilizer on the soil by using reliable and sensitive soil quality indicators. The activity of soil enzymes can indicate the degree of decomposition of soil organic matter (SOM), as enzymes integrate information about the microbial state and physicochemical conditions of the soil.

The aim of the study was to assess the impact of the applied innovative fertilizer mixtures on the enzymatic activity in the contaminated soil. We conducted analyzes of the activity of dehydrogenases, ureases, arylsulfatases as well as acid and alkaline phosphatases. The geometric mean of enzyme activities (GMea) was used to estimate soil quality index. In order to assess the impact of the use of mineral-organic mixtures on enzymatic activity in the soil (with evaluated concentration of cadmium, lead, and zinc), a pot experiment was carried out in a vegetation hall with the use of maize as a test plant. The applied fertilizer mixtures contained NPK mineral components with the addition of zeolite composites and lignite or leonardite as a source of external organic matter. In our research, the highest GMea value was found in the treatment with the addition of 3% zeolite-vermiculite composite and 3% leonardite. Similar values were also obtained in soils with the addition of 9% of zeolite-vermiculite composite and 6% of lignite, as well as 9% of zeolite-vermiculite composite and 6% of leonardite. The conducted studies indicate that the addition of zeolite-vermiculite composite contributed to a greater extent to the improvement of enzymatic activity in the soil than the addition of zeolite-carbon composite.

Keywords: zeolite, external organic matter, contaminated soil, enzyme activity, soil quality index

Acknowledgment: The “Fly ash as the precursors of functionalized materials for applications in environmental engineering, civil engineering and agriculture” no. POIR.04.04.00-00-14E6/18-00 project is carried out within the TEAM-NET programme of the Foundation for Polish Science co-financed by the European Union under the European Regional Development Fund.

Effects of light intensity and spectral composition on growth and selected compounds of garden cress

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Summary

The light environment significantly affects growth, but also the quality parameters of plants, including the content of biologically active substances. In addition to light intensity, the spectral composition of light, including invisible components of radiation such as UV radiation and near-infrared radiation, is an important factor. The aim of the work was to analyse the influence of different environments and light sources on germinating plants of cress (*Lepidium sativum* L.). Seeds were germinated on layered filter papers saturated with distilled water, which was replenished once a day for 10 days, during which the plants were kept in a specific light environment. Each of the light environments was subjected to the measurement of light intensity and spectral composition of light in the range of 315-880 nm by the SpectraPEN Mini device, as well as the intensity of UV-A and UV-B radiation by Vernier sensors. The effect of the light intensity factor was evaluated based on a comparison of the influence of direct and diffused natural sunlight. The spectral composition factor was evaluated on variants grown in the environment of an LED panel for growing plants, Osram Fluora, Sylvania and Reptilia fluorescent tubes, as well as diffused sunlight, which did not differ significantly in intensity, but showed significant differences in the spectral composition of the light. We evaluated the influence of the UV factor in the combination of LED panels with Philips UV-B tubes (LED, LED+2UV, LED+4UV, LED+8UV). After 10 days, the plants of each variant were measured and analysed. Accumulation of flavonoids and anthocyanins was assessed non-destructively, with the Multiplex-3 fluorescence system (Force-A, France). The content of chlorophylls and carotenoids was determined spectrophotometrically and evaluated as a unit of fresh weight. An analysis of the fresh and dry weight of plants of individual variants was also carried out. The antioxidant capacity was also evaluated electrochemically, using the e-BQC device (Bioquochem, Spain). The results of the analysis confirmed the highly evident influence of light intensity on the content of flavonoids, carotenoids and antioxidant capacity, while favourable values were observed for the variant exposed to direct sunlight compared to diffused light. Similarly, a significant beneficial effect of low doses of UV radiation (LED+2UV tubes) was demonstrated. However, a further increase in UV led to a reduction in growth, while the quality parameters did not change or worsened. The influence of the spectral composition was also evaluated as evident, but the influence of the spectrum was significantly weaker at the same intensity than the influence of light intensity or the presence of UV radiation. Our results thus confirmed that by choosing a suitable lighting environment we can influence the biological value of fresh plant products.

Keywords: light spectrum, light intensity, UV radiation, flavonoids, pigments

Evaluation of drought tolerance among common bean landraces

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Summary

Drought is becoming one of the major constraints in bean production and the selection of drought-tolerant bean genotypes is one of the most important drought adaptation strategies. Drought-tolerant plants develop various physiological and biochemical adaptations under water deficit conditions. These include changes in water use efficiency, pigment content, osmotic adjustment, and photosynthetic activity. The aim of this study was to evaluate drought tolerance among different bean landraces ('Biser', 'Zelenčec', 'Trešnjevac', and 'Puter'). Plants were grown in river sand and were irrigated with 50 ml of a 1/4-strength Hoagland nutrient solution at the beginning of the experiment. The water content of the control plants was kept at the maximum water capacity of the sand, while in the drought treatment there was no additional irrigation throughout the experiment. During two weeks, five measurements of the stomatal conductance (gsw), boundary layer conductance (gbw), total conductance to water vapor (gtw), transpiration (E), effective quantum yield of PSII photochemistry (ϕ PSII), and electron transport rate (ETR) were measured using the LI -600 poro/fluorometer. Since the onset of drought, 'Biser' showed significantly higher gsw, gtw, E and ETR compared to other landraces. These results suggest that 'Biser' can maintain normal physiological functions and survive longer under water deficit conditions.

Keywords: common bean, drought, stomatal conductance

Conservation tillage effects on soil health in maize production

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Summary

Conservation tillage is a type of land management that aims to minimize the frequency or intensity of tillage operations in an effort to protecting the soil by managing crop residue and limiting soil disturbance with minimum or no-tillage. Conservation tillage methods improve soil health and productivity, reduce runoff, increase soil carbon sequestration and limit the extent of erosion and also promote certain economic and environmental benefits as decrease in carbon dioxide and greenhouse gas emissions, less reliance on farm machinery and equipment, and an overall reduction in fuel and labor costs. Changes in the soil health status resulting in a diminished capacity of the ecosystem to provide goods and services for its beneficiaries, are define as soil degradation. The field experiment with different conservation tillage systems in maize production was conducted in 2021 in Čačinci and Križevci. Tillage treatments were: ST (plowing), CTD (conservation deep, loosening up to 30 cm with a minimum of 30% of crop residues) and CTS (conservation shallow, tillage up to 10 cm with a minimum 50% of crop residues). The following parameters were investigated: bulk density (ρ_v), packing density (PD), soil compaction (by penetrometer), soil organic matter (SOM), soil organic carbon (SOC), soil respiration, estimation of N-mineralization and C/N ratio. After the first year of research, a large heterogeneity of the researched parameters between the tillage treatments was recorded.

Keywords: conservation tillage, soil degradation, soil health.

This work has been fully supported by Croatian Science Foundation under the project “Assessment of conservation soil tillage as advanced methods for crop production and prevention of soil degradation – ACTIVEsoil” (IP-2020-02-2647)

The effect of sewage sludge application on soil chemical properties during the cultivation of energy crop Virginia Mallow (*Sida hermaphrodita* L.)

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Summary

The aim of this research was to determine the effect of application of the 1.66 t ha⁻¹ dry matter of sewage sludge allowed by the Croatian ordinance on chemical properties of soil after two years. After the harvest of Virginian mallow, no statistically significant influence was recorded on any of the observed properties. Regardless of the determined amounts of heavy metals in the soil depending on fertilization treatments, it was determined that the values for cadmium, chromium, copper, mercury, nickel, lead, zinc, molybdenum and arsenic are below the maximum allowed concentration according to the Ordinance [Protection of agricultural land from pollution (NN 71/2019)]. This is in accordance with one of the hypotheses, according to which there will be no significant impact on the properties of the soil, especially with regard to toxic heavy metals.

Keywords: chemical properties, heavy metals, sewage sludge, pollution, Virginia Mallow

Effects of fallow and green manure on soil characteristics and yield of spring crops

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Summary

Several European Union policies (e.g. Green Deal, Climate law, Biodiversity Strategy 2030, Farm to Fork) have a goal to transform the agricultural systems and one of the goal is fertilizers reduction. Nature based solutions (e.g. green manure) can help in restoring soil quality for enhanced agricultural productivity and sustainability. A two-year research (on the Dalibor Jurina family farm in Veliki Zdenci, Bjelovar-Bilogora County) was conducted to test real fallow and green fallow on the improvement of the soil quality. Experiment is conducted in random block layout with four treatments [I. black fallow; II. Black fallow + green manure; III. Crops (2021-corn & 2022-soybean) + green manure; IV. crops (2021-corn & 2022-soybean) without green manure] on plots of 10 x 35 m in four repetitions on Pseudogley on the plain in semi-humid conditions. Results confirmed that green manure affects changes in the soil reaction, the stability of structural aggregates, and C/N ratio. Soil compaction is reduced in green manure plots over a period of two years (autumn 2020 - autumn 2022). Moreover, there is no correlation between green manure and maize yield in the first year of the study. Results reveal that the maize yield was lower in the treatment where green manure was applied. In the second year of the study, the yield of soybean was higher in the treatment with green manure. It is necessary to continue research in order to determine changes in physical and chemical properties and to observe the influence of green manure on them, as well as on the yield of cultivated crops.

Keywords: green manure, physical and chemical properties, corn and soybean yield

Two-year survey of the presence of aflatoxigenic and ochratoxigenic fungi in an experimental field in Osijek

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Summary

Mycotoxins are low-molecular-weight secondary metabolites synthesized by some filamentous fungi, toxic to animals and humans. As a contaminant, they can be found on different feed and foodstuffs, especially on cereal grains. Therefore, they pose a major problem worldwide. Within the project “Food production of biocomposites and cereals in a circular economy”, monitoring the occurrence of ochratoxigenic and aflatoxigenic fungi was carried out during a two-year survey in the experimental field in Osijek. Air sampling in the field was conducted during May, July and October of 2021 and 2022. Sampling was performed using an Air Sampler, which was adjusted so that the airflow through the Petri dishes for sampling was 100 L min^{-1} . Two nutrient media; Potatoe Dextrose Agar and Sabouraud Dextrose Agar, were used to isolate conidia from as many species of fungi present in the air as possible. Mycotoxigenic airborne molds in maize, wheat, and barley plantations, as well as in maize cobs were isolated and morphologically determined. Moreover, an assay of the ability of potential mycotoxigenic fungi to synthesize ochratoxin A and aflatoxin was performed. The results of the research are in accordance with the preliminary results of the first-year study, which showed the presence of both aflatoxigenic and ochratoxigenic fungi in the field with a percentage of occurrence of less than 10%. Analysis of the presence of aflatoxins and ochratoxin A in the harvested cereals is in the progress. Since the risk of contamination is present, it is necessary to continue monitoring in order to implement preventive control measures for aflatoxin and ochratoxin A contamination.

Keywords: aflatoxigenic fungi, cereals, food safety, mycotoxigenic contamination, ochratoxigenic fungi,

Vermicomposting of biofortified harvest residues into high-quality organic fertilizers

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Summary

The management of organic matter is increasingly important in all agroecosystems, both for the sustainability of agricultural production and for environmental protection. In doing so, it is very important to produce quality organic fertilizers with as little loss of organic C and N as possible, for example by vermicomposting with the use of earthworms. At the same time, in the world, more than a third of the population is exposed to a certain form of malnutrition due to the insufficient content of microelements in food, which can be mitigated by agronomic biofortification, i.e. the application of microelements during crop cultivation. The goal of biofortification is the production of enriched grain, while significant amounts of crop residues, also enriched with microelements, remain on the arable land. The aim of this research was to determine the possibility of using harvest residues after biofortification in the production of vermicompost enriched with Se and Zn. A two-year study of agronomic biofortification of corn and soybean was carried out by individual foliar applications of Se (10, 20 or 30 g/ha) or Zn (3 or 6 kg/ha). In addition to the successful enrichment of soybean and corn grains, a large increase in the concentrations of Zn and Se in the harvest residues was also determined. The highest concentrations of Zn and Se were determined in the leaves of soybean (247-620 mg/kg Zn and 315-1,788 µg/kg Se) and corn (52-198 Zn and 376- 2,056 Se), and significantly less in stems of soybean (16-56 mg/kg Zn and 57-658 ug/kg Se) and corn (35-58 mg/kg Zn and 98-401 ug/kg Se), depending on the biofortification treatment. Biofortified harvest residues were used to prepare different mixtures (control, Zn-enriched, Se-enriched mixture) for vermicomposting using horse manure, grape pomace and soybean and corn harvest residues. Vermicomposting was carried out in pots with 5 kg of mixture, with the addition of 50 earthworms *Eisenia andrei* to each pot (initial weight 10-16 g per pot). After 120 days 35% more Zn (68,6 vs. 50,8 mg/kg) and 50% more Se (145 vs. 97 µg/kg) were found in vermicomposts from enriched crop residues than in control treatments. Higher concentrations of Zn and Se increase the fertilizing value of vermicompost for soils poor in available Zn or Se fractions. At the same time, there is no fear of the toxic effects of Zn and Se, since a positive reaction of earthworms to biofortified harvest residues was determined, as compared to the control treatment. The number and weight of earthworms compared to the initial values were increased (number 150% for Zn and 98.5% for Se; weight 34.1% for Zn and 9.1% for Se), while in the control treatment the number was increased only by 50% with an unchanged weight of earthworms.

Keywords: selenium, zinc, earthworm, soybean, corn

Impact of microbial bioagents application and mineral fertilization on wheat yield

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Summary

Sustainable agricultural production, in addition to reducing the use of mineral fertilizers and optimizing the use of organic fertilizers, also implies the use of microbial bioagents with the aim of achieving the target yield while simultaneously protecting the environment and soil fertility. However, the use of microbial bioagents must be in accordance with the soil properties, the needs of the crops and the characteristics of the microbes. The aim of this research was to determine the influence of microbial bioagents on winter wheat yield with optimal and reduced fertilization with mineral fertilizers. The field experiment was set up in the fall of 2021 after maize on Trnava locality on acid poor soil. In addition to the two levels of fertilization, optimal and reduced, the experimental treatments were 4 different microbial bioagents: MB1. VAM + *Azotobacter* + *Azospirillum*, MB2. MB1 + *Bacillus* + *Pseudomonas*, MB3. MB1 + *Bacillus* + *Trichoderma* + *Beauveria*, MB4. MB2 + MB3 and 4 different application treatments (1. control without application, 2. application in soil, 3. seed treatment, 4. combination 2+3) in four replications. There were 128 plots (2 × 4 × 4 × 4) and a total experimental area of 6.4 ha. The yield of wheat was 8.5 t/ha in average and optimal fertilization (8.7 t/ha) resulted in a yield increase of 2.5 t/ha comparing reduced fertilization (6.2 t/ha) without the use of microbial bioagents. On average, the use of microbial agents increased the yield by 1.4 t/ha, which is an increase of 18.4%. The average yield of wheat after optimal fertilization and application of agents was 9.6 t/ha, with almost no differences between application methods, with a 10.5% increase in yield compared to optimal fertilization without bioagents. However, in the case of reduced fertilization, the increase with the application of bioagents compared to the control was 29.4%, and the highest yield with reduced fertilization was achieved with seed treatment. But the use of bioagents decreased the difference between reduced and optimal fertilization from 2.5 t/ha on control treatments to 1.8 (seed treatment and application in soil), 1.6 (application in soil) and 1.3 (seed treatment). The conclusion is that microbial bioagents can additionally increase the yield of wheat on poor soil both with reduced and optimal mineral fertilization, with a relatively higher effect on yield increasing after reduced fertilization.

Keywords: *Azotobacter*, *Pseudomonas*, mycorrhizae, acid soil, reduced fertilization

Maize yield after microbial bioagents application under different fertilization

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Summary

For decades, the intensive cultivation of field crops meant the use of mineral fertilizers in order to achieve the target yield and preserve soil fertility. However, the development of agriculture in the direction of sustainable management includes the reduction of the mineral fertilizer application by more intensive use of organic fertilizers and microbial bioagents. Therefore, it is very important to determine the influence of microbial bioagents on the yield in cultivation with different levels of fertilization. The aim of this research was to determine the influence of microbial bioagents on corn yield with moderate and reduced fertilization with mineral fertilizers. Maize (FAO 390) experiment was set up in 2021 on Trnava locality on acid poor soil with a total of 128 experimental plots ($A_2 \times B_4 \times C_4 \times 4$) and a total experimental area of 6.4 ha. The main treatments of the experiment were moderate (A_1) and even more reduced (A_2) mineral fertilization with 4 different microbial bioagents: B1 (VAM + *Azotobacter* + *Azospirillum*), B2 (B1 + *Bacillus* + *Pseudomonas*), B3 (B1 + *Bacillus* + *Trichoderma* + *Beauveria*), B4 (B2+B3). All four microbial agents were applied in 4 different treatments (C1. control without application, C2. application in soil, C3. seed treatment, C4. application in soil with seed treatment) in four replications. The yield of maize was very low (6.18 t/ha in average), but 12.3% higher after moderate comparing to reduced fertilization. On average, the use of microbial agents increased the yield by 0.9 t/ha, which is an increase of 17.1-17.8%. In total, the combination of seed treatment and application of bioagents in the soil was the most successful with an increase in yield of 1.2-1.4 t/ha (23.6-24.9%), followed by the seed treatment (1.1-1.34 t/ha). The most significant individual effects were determined for seed treatment with bioagents B2 (increase of 2.85 t/ha, 50.5%) and B4 (increase of 2.28 t/ha, 40.4%). The conclusion is that microbial bioagents can effectively replace a part of mineral fertilizers even on poor acidic soils where the most effective is a seed treatment with bacteria and mycorrhizae.

Keywords: *Azotobacter*, *Pseudomonas*, mycorrhizae, acid soil, seed treatment

Influence of less permeable subsurface horizon of Pseudogley on grapevine root distribution

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Summary

Pseudogley is the soil that is characterized by the presence of a less permeable subsurface horizon that affects the percolation of water and restrictions in root growth. The aim of this work was to determine the influence of a less permeable subsurface (Btg) horizon on the distribution of grapevine (*Vitis vinifera* L.) roots. Soil profiles were dug at three positions (top (P1), middle (P2), bottom (P3)) on the slope, where the root distribution was determined up to 100 cm deep, in the row and in the inter-row area of the vineyard (along and perpendicular to the row of vines). The analysis of root distribution was carried out using a 1 m² grid with hole diameters of 10 × 10 cm, where the number of roots of different diameters (> 5; 5 – 2; 2 – 0,2; < 0,2 mm) was determined in each of the holes. In general, in each of the slope positions, a greater number of roots was detected in the row compared to the inter-row space of the vineyard. The surface parts of the Ap horizon (0 – 20 cm) at each of the positions were extremely rooted containing a greater number of smaller (< 0,2 mm) roots. An average of 40 – 50% of the total number of roots at a depth of 0 – 100 cm was recorded in the inter-row part of the vineyard in the surface 0 – 20 cm, while 65 – 80% of the number of roots was recorded up to a depth of 0 – 40 cm. In a row, up to a depth of 0 – 20 cm, the presence of roots ranges from 40 – 50% and up to a depth of 0 – 40 cm, the presence of roots is 60 – 70%. Therefore, the results of the research indicate the fact that the majority of vine roots develop above the less permeable horizon.

Keywords: slope positions, root distribution, less permeable soil horizon

Transformations of organic matter in soil after application of mineral-organic mixtures with the addition of zeolite-vermiculite composite

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Summary

Due to the indisputable significance of humus in many biochemical processes as well as its increasing deficit particularly in light soils, alternative sources of substrates for the reproduction of this constituent should be sought. Humic substances are a reservoir of organic carbon in the soil, play a very important role in the immobilization of pollutants, increase water retention, and indirectly affect the growth and yield of plants. Undoubtedly, one of the activities aimed at increasing soil organic matter is the application of various organic materials, including materials of waste origin. The aim of this study was to evaluate the effect of the addition of synthetic zeolite-vermiculite composite in combination with lignite and leonardite on the qualitative and quantitative parameters of humic compounds in contaminated soil. In the obtained soil samples after the second year of pot experiment, the content of soil organic carbon was determined using the Tiurin method and the fractional composition of humic compounds using the Kononowa and Bielszikowa method. The content of non-hydrolyzing carbon in the soil was significantly increased as a result of treatments with the addition of mineral-organic mixtures, which indicates a greater stabilization of humus compounds and, at the same time, lower CO₂ emissions. Changes in the carbon content in humic and fulvic acids after fertilization altered the CHA:CFA ratio value. Applied fertilizer mixtures increased the content of organic carbon in the soil. Research on organic substances is important not only from the cognitive point of view, but also in terms of assessing the fertility or degree of soil degradation.

Keywords: soil organic matter; fertilizers; zeolite-composite; humic compounds;

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Influence of the plant host on the variability of the invasive Mediterranean fruit fly

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Summary

The Mediterranean fruit fly (*Ceratitis capitata*, Wiedemann) is an invasive and quarantine pest, infesting more than 360 different plant species in tropical, subtropical, and Mediterranean climates. It originated in warm regions of Africa and has been causing significant damage in Croatia since the 1950s. Initially, the Mediterranean fruit fly was found exclusively in citrus growing areas and posed a greater threat especially to growers in the Neretva region. Today, the pest is widespread along the entire Adriatic coast and is sporadically found on the mainland as well. In Croatia, it causes the greatest damage and economic losses in the cultivation of mandarins, but its presence has also been noted in the cultivation of peaches and figs, which it also severely damages. The aim of this work was to determine the variability of populations of the Mediterranean fruit fly based on wing shape, from different hosts (peach, fig and mandarin) from different cultivation areas in the Republic of Croatia. Through variability and assessment of phenotypic plasticity, which determines the invasive character of the pest, geometric morphometrics methods were to be used to determine the potential for adaptation and future spread of the pest to new areas and new plant hosts. The study tested 10 different populations from peach, fig, and mandarine, as well as a sterile population grown under laboratory conditions. It was found that the hosts in which the Mediterranean fruit fly develops influence the shape of the wings, i.e., condition their variability. In addition, high phenotypic plasticity was found, which is a prerequisite for the invasiveness and spread of the Mediterranean fruit fly to new areas and new hosts in the future.

Keywords: medfly, wing shape, adaptability, invasiveness

Isotopic signatures of nitrogen in selected soils of Croatia

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Summary

Nitrogen (N) is an essential element for all living beings on Earth. In nature nitrogen exists as a mixture of two stable isotopes (¹⁴N and ¹⁵N) and by its composition and proportion the environment and soil evolution can be characterized. The natural nitrogen isotopic signature ($\delta^{15}\text{N}$) in the terrestrial ecosystems is conditioned by the complexity of the nitrogen cycle and includes all its transformations and degrees of isotopic fractionation. The aim of this research was to determine the variability of total nitrogen and $\delta^{15}\text{N}$ isotopes (isotopic signature of nitrogen) in selected soils of Croatia due to different land use (arable land, meadow, forest, orchard, ski slope, urban area). The research included the analysis of 27 archived soil samples collected in the period 2005-2020 as part of scientific work of employees of Department of General Agronomy (University of Zagreb Faculty of Agriculture). Soil samples were collected from eleven locations in Croatia (Vukovar, Potok, Molve, Zagreb, Žumberak-Samoborsko gorje, Lepoglava, Karlovac, Gospić, Vodnjan, Pag, Mljet). Total nitrogen was determined by the dry combustion method, and $\delta^{15}\text{N}$ isotope values by the EA-IRMS method. Laboratory analyses were conducted in 2021. The research results indicate a range of $\delta^{15}\text{N}$ values from 0.0 ‰ to +7.19 ‰, and total nitrogen content from 0.084 % to 0.991 %. In most of the observed soils (21 samples), the isotopic nitrogen signature indicates the organic origin of nitrogen in the soil (3.81 ‰ to 7.19 ‰), in three soil samples (two meadows soils and a ski slope soil) the nitrogen originates from mineral nitrogen fertilizers (2.20 ‰ to 3.20 ‰), while in three forest soil samples nitrogen atmospheric deposition contributed to its origin (0.00 ‰ to 3.24 ‰).

Keywords: arable land, meadow, forest, orchard, $\delta^{15}\text{N}$

The necessity to diversify regional labeling of biodynamic products

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Summary

The purpose of this paper is to determine opinions and attitudes on the necessity of diversification of food products that are produced according to the standards of biodynamic agricultural practice. In the Republic of Croatia, biodynamic products on their packaging are marked only with the label of eco products, although according to their principles and production methods they represent a greatly different over standard to the already defined eco-label (standard). According to research dealing with the so-called “green products”, it was concluded that products marked with an eco and / or biodynamic sign represent a trusted good.

In this paper, the scientific approach defined the observed problem and conducted research on the necessity of diversification of labeling of biodynamic products. The subject of the research was chosen due to many years of experience and noticing the lack that there is no quality producer-media-consumer communication process that would satisfy the purpose of characterization and diversification of specific biodynamic products on the Croatian market.

Empirical research was conducted through a questionnaire using the created Google form with 24 questions structured in three parts. The study included 194 subjects of both sexes, aged 19 to 72 years. The research found that without a recognizable label on the packaging of biodynamic products, manufacturers and consumers face the problem of identifying these high-quality products on the market on a daily basis.

The research found that without a recognizable label on the packaging of biodynamic products, producers and consumers face the problem of identifying these high-quality products on the market on a daily basis. Therefore, the identification of key parameters that contributed to the creation and design of an original, innovative, relevant and authentic graphic label for the packaging of biodynamic products was initiated in order to propose adequate solutions for the standardization of labelling of biodynamic products at the national level.

Keywords: biodynamic products, diversification of biodynamic products, labelling

Extended-spectrum β -lactamase producing *Enterobacterales* from hospital wastewater in Croatia

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Summary

Hospital wastewater is a source for transmission of extended-spectrum β -lactamase (ESBL)-producing *Enterobacterales* into the municipal sewage system and further into the environment. Here, we describe the occurrence of ESBL-producing enterobacteria, and their resistance genes from wastewater of two major hospitals in Zagreb, Croatia. Chromogenic medium RAPID'E. coli2 agar (Bio-Rad) supplemented with cefotaxime (4 mg/L) was used for the isolation of presumptive ESBL-producing enterobacteria. Identification was done with MALDI-TOF MS. Antibiotic susceptibility profiles and ESBL production was assessed. ESBL and carbapenemase genes were detected using PCR. Out of 100 presumptive ESBL-producing enterobacteria, the most dominant taxa were *Escherichia coli* (48 %), *Citrobacter* spp. (14 %), *Enterobacter* spp. (13 %) and *Klebsiella* spp. (8 %). The 69 isolates were ESBL-producing enterobacteria with multi-drug resistance. The predominant ESBL genes detected with PCR were bla_{TEM} (95 %) and $bla_{CTX-M-1}$ (87 %). The bla_{SHV} and bla_{GES-7} were detected sporadically. Moreover, more than 50 % of ESBL-producing isolates were resistant to carbapenems, and bla_{KPC-2} and bla_{OXA-48} genes were the most detected carbapenemase genes. The occurrence of ESBL-producing isolates indicates that hospital wastewater is the source of multi-drug resistant enterobacteria and is evidence of a need for pretreatment of hospital wastewater before discharge into the municipal sewer system.

Keywords: *Enterobacterales*, extended-spectrum β -lactamase, carbapenemase, hospital wastewater

An insight into phenological dynamics of karst pasture after controlled burning – a case study from Dinara Mountains

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Summary

After the introduction of the EU's Common Agricultural Policy, a new interest in livestock-keeping has been sparked among the local population of Dinara Mountains. But the pastures, which have been traditionally maintained by mowing, grazing and semi-controlled burning, are largely in succession after the abandonment of the traditional practice of transhumance in the 20th century. In February 2021 controlled burning has been conducted on an overgrown karst pasture in Vrdovalo as part of the Dinara back to LIFE project. The aim of the study was to compare the phenology of Vrdovalo grassland before and after burning by analyzing NDVI dynamics (a proxy for phenology) during the last five years. NDVI was obtained from Sentinel-2 images dating from January 2018 to September 2022 for five points of each land class detected on the grassland. The average monthly NDVI was calculated per class for every year. One-way ANOVA was carried out but no significant differences were found. However, a shift in the peak of NDVI (peak of vegetation season) has been observed in all land classes after 2021 with the peak being recorded in June instead of July and August as in previous years. The NDVIs recorded in June 2021 and 2022 were also the highest during the study period. It's unclear if the changes in the peak of vegetation season were linked to the burning but given the current results, we cannot conclude that the controlled burning impacted the phenology of Vrdovalo grassland. Further research is needed.

Keywords: controlled burning, Dinara, karst grassland, NDVI, phenology

Acknowledgments: This study was conducted as a part of the project „Dinara back to LIFE – Management planning and restoration of Dinara dry grasslands to save biodiversity and support sustainable development“ (LIFE 18 NAT/HR/000847). The project is co-funded by the Environmental Protection and Energy Efficiency Fund.

Platinum, palladium and rhodium in Zagreb air, soil and vegetation

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Summary

Platinum (Pt), palladium (Pd) and rhodium (Rh) are used in various branches of industry, from electronics to dentistry, jewelry and medicine, to the automotive industry where their greatest application is in the production of automotive catalytic converters that serve to reduce toxic emissions from motor vehicles. However, hot exhaust gases that pass through the automotive catalyst cause emissions of Pt, Pd and Rh into the environment. The increasing use of automotive converters has led to an increase in the concentrations of mentioned metals in the environment and thus to a higher bioavailability, which potentially raises concerns about the insufficiently researched environmental impact of Pt, Pd and Rh on human health. The aim of this research was to determine the concentrations of platinum, palladium and rhodium and their seasonal and spatial distribution in air, soil and vegetation. Weekly samples of particulate matter with an aerodynamic diameter of less than 10 µm (PM₁₀ fraction) were collected throughout the 2020 at three urban monitoring stations in Zagreb. Vegetation and soil samples were collected during two seasons, in spring and autumn; two samples of aboveground plant material (*Plantago Lanceolata* L. and *Dactylis glomerata* L.) and two soil samples (at 2 depths: 0-5 cm and 5-10 cm) from the close proximity to existing air quality monitoring stations. Samples were prepared by microwave digestion in acid under high pressure and temperature, and analyzed by inductively coupled plasma mass spectrometry (ICP MS). The results show significantly higher concentrations of palladium in soil, air and vegetation than rhodium and platinum. Seasonal, spatial, and for soil additional depth differences were observed for platinum, palladium and rhodium. *Plantago Lanceolata* L. shows a higher content of measured metals than *Dactylis glomerata* L. and also has indicator properties, especially for palladium.

Keywords: ICP MS, microwave digestion, PM₁₀, *Plantago Lanceolata* L., *Dactylis glomerata* L.

Effect of earthworms on the decomposition of TPH compounds in sewage sludge compost

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Summary

The total petroleum hydrocarbon (TPH) content of sewage sludge and sewage sludge compost can be a potential risk for soil health during their agricultural utilization. Treatment of sewage sludge by vermicomposting causes more intense decomposition and transformation processes compared to composting. Thus, it can be assumed that it is also effective in reducing TPH compounds. In a model experiment, the effect of earthworm activity was examined on the degradation of TPH compounds in sewage sludge-based compost. Gearbox oil was mixed in loads of 0, 0.5, 1, 2 and 3 g/kg D.M. with commercial compost in pots containing 1 kg wet compost. The total number of pots was 5 loads x 2 earthworms (present/absent) x 3 replicates = 30 pots. After two weeks of incubation, earthworms (*Eisenia fetida*) were placed in the appropriate pots (30 pcs/pot). The pots were covered but not sealed from air and were kept in room temperature. The duration of the experiment was 3 months from the time of earthworm installation. The TPH had no toxic effect on earthworms. At the end of the experiment each animal was alive however, no signs of reproduction were seen. As a result of the TPH decomposition, the organic nitrogen of the compost was mineralized, which doubled the NO₃-N concentration in the material regardless of the presence of earthworms. The amount of organic matter was statistically the same at all TPH loads in treatments with and without earthworms. The organic matter stability was higher in the treatments without earthworms. The final TPH concentration was statistically the same at all loads with or without earthworms. This was probably caused by the fact that during the 3-month period of the experiment, the microbial community of the compost degraded TPH compounds regardless of the presence of earthworms.

Keywords: vermicomposting, organic contaminant, waste treatment

This work was funded by the National Research Development and Innovation Office, project number: 2021-1.2.4-TÉT-2021-00035, entitled “Macronutrients and micropollutants in the soil-plant system: from rational nutrient supply to soil contamination.”

In-vessel composting of grape pomace

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Summary

Composting is defined as a process of aerobic degradation of organic waste by microorganisms what results with a stabilized, mature, hygienic product, free of pathogens and rich in humic substances. Thanking into account that winemaking process produces a wide range of solid and liquid by-products including grape marc or grape pomace, wine lees, spent filter cakes, vinasses, and winery wastewater that must be properly treated, disposed of, or reused to prevent adverse environmental effects, composting could be effective way to reduce the wine industry negative environment impacts. Compost, the final product, can improve the soil structure, helps maintain a neutral pH and enhances plant growth. In this work a laboratory composting of grape pomace was performed in a closed thermally insulated column reactor ($V = 5$ L) with continuous aeration during 20 days. The temperature changes, moisture content, pH-value, and C/N ratio in the composting pile were investigated. The results suggested that adiabatic conditions enhance the grape pomace composting process.

Keywords: grape pomace, adiabatic composting, food waste valorization

Acknowledgment: The work was supported by the European Union through the European regional development fund, Competitiveness and Cohesion 2014–2020 (KK.01.1.1.07.0007.).

Correlations between fertilizer use and potentially toxic element content in Hungarian soils

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Summary

Mineral fertilizers serve as sources of trace elements for cropped plants. However, by unreasonable doses and repeated fertilizer application the total potentially toxic element (PTE) concentration of soils might increase. The main objective of this research was to investigate possible PTE load originating from fertilizer use in the Hungarian test areas. We looked for detectable correlations between different levels of fertilizer uses and the concentration of PTEs in the soil.

The analysed soil samples (n=129) resulted from a nation-wide survey. Bulk soil samples (0-30cm) were taken in 2011. Aqua regia soluble „total” PTE content was measured by ICP-OES. Fertilizer output data (N, P₂O₅ and K₂O active ingredient doses) of the three years prior to sampling were known from a preliminary survey.

According to the results, the sampling sites with no fertilizer output had a lower concentration of most of the PTEs than those with either N, P or K output. Above 100 kg/ha NPK active ingredient doses, however, a tendency of increasing fertilizer doses causing higher PTE concentrations could not be detected. Besides, huge differences could be observed among the elements. The concentration of PTEs did not show statistically provable increase as the result of present fertilizer application. It can be assumed that toxic element content of Hungarian soils is mainly influenced by other factors rather than agricultural load.

Keywords: mineral fertilizer, potentially toxic elements, bulk soil samples, pollution

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Multispectral assessment of aluminium toxicity on corn

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Summary

Aluminum (Al) is the most abundant metal constituent of the Earth's crust with an average amount of 81000 mg kg⁻¹ in the lithosphere. Under acid conditions, soil aluminum content affects crop growth; several studies have found aluminum toxicity in crops. Aluminum toxicity primarily affects root growth, but also causes changes in the carbon dioxide assimilation rate of plants, the chlorophyll content of leaves, and affects a number of physiologically important enzyme activity. The symptoms of the aluminum toxicity are: stunning, necrosis of leaf tips, small, dark leaves and purpling of stems, and root inhibition. The aim of this study was to investigate the effect of aluminum toxicity of the multispectral characteristics of a Hungarian fodder corn (*Zea mays*, Armagnac hybrid). The experiment was set up under control conditions in growing chamber. The treatments were: control (0 μM); 50 μM, 100 μM, 200 μM and 400 μM AlCl₃ L⁻¹. The pH of the solutions was set to 4, because the aluminum toxicity appears in acid soils above pH 5. The multispectral parameters were measured two times per week using by the Crop Reporter. This experiment extends the study of the effects of aluminum toxicity on the plant physiological parameters of maize. Aluminum toxicity affected plant corn reflectance parameters which was accompanied by changes in vegetation indices correlated with pigment content.

Keywords: corn, aluminum stress, vegetation indices, reflectance, multispectral assessment

Some soil safety aspects of organic fertilizer utilisation

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Summary

Maintaining and improving the good condition and fertility of the soil requires special attention from the aspect of environmental safety. During our research, we studied the environmental effects and agricultural applicability of organic fertilizers from an ecotoxicological point of view.

In a mesocosm experiment, we investigated the environmental risk of four organic fertilizers (sewage sludge compost, digested sewage sludge, littered cattle manure and pig slurry) on calcareous chernozem and sandy soils with the help of two test organisms. The organic materials mixed in each treatment were applied in 4 doses (0, 85, 170 and 340 kg N/ha) in 3 replicates. For the ecotoxicological study, we chose a plant (*Sinapis alba* root growth inhibition) and an animal test (*Collembola* (*Folsomia candida*) acute toxicity test) according to the OECD standards.

In the case of the chernozem soil all treatments had a stimulating effect on root growth compared to the control with the exception of pig slurry which resulted in an inhibitory effect at the highest dose. On the sandy soil, the highest dose of the fertilizers increased the length of the roots by almost five times. Based on the number of germinated seeds, even the highest provocative doses did not inhibit germination in any of the treatments.

The *Collembola* test did not indicate any negative effects in any of the treatments

Based on our results, in the case of the four tested fertilizers, the short-term exposure did not cause an inhibitory effect on plant root growth, with one exception, nor did it affect the lethality of the test animals.

Keywords: soil safety, environmental risk, ecotoxicology, organic fertilizer, organic waste

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Carbapenem resistance in Croatian municipal wastewater

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Summary

Bacterial carbapenem resistance, especially among *Enterobacteriaceae*, is a major public health problem and wastewater treatment plants (WWTPs) can promote its spread in the environment. In this work we describe the occurrence of carbapenem-resistant coliforms and carbapenemase genes and the characteristics of carbapenemase-producing enterobacteria from municipal wastewater in Croatia. Conventional culturing was performed to monitor coliform bacteria and quantitative PCR to monitor 5 carbapenemase (CP) genes in the influent and effluent of 7 Croatian WWTPs in two seasons. Carbapenem-resistant enterobacteria were isolated on CHROMagar mSuperCARBA plates from the effluent of the largest WWTP (Zagreb) and identified by MALDI-TOFF. Antimicrobial susceptibility profiles and carbapenemase production were assessed phenotypically. Detection of CP genes was performed by PCR and sequencing of selected amplicons or by whole genome sequencing (WGS). Molecular epidemiology was assessed by PFGE and WGS. In general, levels of total and carbapenem-resistant coliforms were significantly reduced but not eliminated by conventional treatment in most WWTPs. CP genes, *blaKPC-3*, *blaNDM* and *blaOXA-48*-like, were sporadically detected, while *blaIMP* and *blaVIM* were frequently enriched during treatment and were influenced by specific features of the WWTPs. A total of 200 carbapenem-resistant enterobacteria were isolated from Zagreb WWTP effluent. Most isolates (144/200) produced carbapenemases and were confirmed as *Enterobacteriaceae*, with the predominant taxa identified as *Klebsiella* (33%), *Enterobacter* (24%), and *Citrobacter* (25%). All *Klebsiella* isolates were multidrug-resistant, of which 6% were pandrug-resistant. Most *Klebsiella* isolates were grouped into 8 pulsotypes; among these, two were predominant and belonged to a novel *Klebsiella* lineages, ST1697 and ST3590, which may be a hybrid of the recombination of the *Klebsiella quasipneumoniae* and *Klebsiella pneumoniae* genomes. The predominant CP genes among these isolates were *blaOXA-48*, *blaNDM-1* and *blaKPC-2*, whereas *blaVIM-1* and *blaIMP-13* were rarely detected. 40% of *Klebsiella* isolates were resistant to colistin, and a novel plasmid-encoded *mcr-4.3* gene was detected for the first time in pandrug-resistant *Klebsiella pneumoniae*.

Our results suggest that improvements in WWTP technologies are needed to minimize the risk of environmental contamination with key opportunistic enteric pathogens and CP genes and the resulting public health impact.

Keywords: carbapenem, resistance, wastewater, enterobacteria

Reliability of visual diagnostics in determining iron deficiency on apple leaves in comparison with chemical analysis

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Summary

The assessment of nutrient deficiencies on plant leaves and fruits, taking into account visual symptoms specifically, in plant nutrition is known as visual diagnostics. It is the initial, but not the only step in determining the nutritional status of the plant. Visual diagnostics can serve as a method to indicate nutrient imbalance in the plant, but it should not be a reference point for drawing conclusions and accordingly making recommendations for fertilization. In this research, the iron status of apple (*Malus domestica*) plants, whose deficiency characteristic is interveinal chlorosis with occurrence on young leaves, was examined by visual diagnostics. The aim was to determine the reliability of visual diagnostics by comparison with the results of chemical analysis of apple leaves. Samples of young, middle-aged, and old leaves on three shoots were taken from an intensive apple orchard near the city of Velika Gorica (Ribnica). After determination of iron deficiency by visual diagnostics on young and middle-aged leaves, chemical analysis showed its lack on all leaves, regardless of age. All determined values ranged from 30.1 to 57.5 mg/kg, which is at the lower limit of optimal values (40-400 mg/kg) for apple according to Bergmann (1992). The visual diagnostics used in this case to assess the iron status in apple leaves proved to be correct and reliable.

Keywords: visual symptoms, nutrients, young leaves

Simultaneous encapsulation of two antimicrobial chemical agents in alginate/chitosan microparticles

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Summary

Pathogenic microorganisms, including fungi, oomycetes, bacteria, viruses, and viroids, constitute a serious threat to agriculture worldwide and are one of the biggest threats to global commercial crop and food production resulting in significantly increased cost of production, reduced crop establishment and productivity. One of the main challenges of modern agriculture is the preparation of new agroecological formulations for the protection of plant crops based on silver and zinc ions. The aim of this investigation is to explore how the method of ionic-gelation encapsulation method can produce biopolymer microcapsules enriched with silver and zinc ions, which could increase the stability and viability of silver and zinc ions over a longer time period. At the optimal point microcapsules enriched with silver and zinc ions were produced and physiochemically characterized (microcapsules size, encapsulation efficiency, loading capacity, particle size). The process of encapsulation in biopolymer microcapsules did not have a negative impact on the properties of the silver and zinc ions and its activity remains very high. Also, the silver and zinc ions release data were fitted to the Korsmeyer-Peppas model and the n exponent indicated that the release mechanism was Fickian. The electrostatic interactions between silver, zinc ions, alginate and chitosan were confirmed by infrared spectroscopy. The obtained results showed that silver and zinc ions could be successfully encapsulated and applied in the process of plant protection against plant pathogens.

Keywords: silver ions, zinc ions microcapsules, plant protection, pathogens

Natural restoration of flora on burned agricultural land - case study Biograd na Moru (Croatia)

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Summary

A neglected arable agricultural area near the town of Vrana near Lake Vrana was burned by an induced fire. The area is located in the transition zone between evergreen (*Fraxino ornī - Quercetum ilicis*) and deciduous vegetation (*Quercō-Carpinetum orientalis*). The agricultural site itself, immediately around the burned area of the experiment, is dominated by Mediterranean maquis with the characteristic species *Pistacia lentiscus* and the monoculture community of *Foeniculum vulgare*. The studied area is divided into three experimental variants (average slope 18%): I. (control - anthropogenic meadow), II. (lower intensity burning, burning 100 kg/m² of straw) and III. (higher intensity burning, burning 100 kg/m² of straw and 150 kg of vine wood). Fifteen rings (diameter 0.2 m²) were established on the studied area, five on each variant. The growth and development of flora and vegetation were observed monthly within 24 months after the fire. All inventoried plant species were identified, herbarized, digitised, and are available in the ZAGR online herbarium (http://herbarium.agr.hr/hr_search.html). Abundant growth of plant taxa from all three functional groups that follow different ecological survival strategies was observed on the control variant: legumes, forbs, and grasses and grass-like plants. The most abundant species included the very species that were recorded within the experimental rings, but also included several others that were not previously recorded in the burned experimental rings (*Plantago lanceolata*, *Urospermum picroides*, *Sisymbrium officinale*, *Silene latifolia ssp. alba*). On the variant with lower fire intensity, the dominance of monocotyledons from the Poaceae family is visible: *Avena sativa*, *Dactylis glomerata*, *Hordeum murinum*, *Elymus repens* and others. On the variant with higher fire intensity, the dominance of plants from the group and family of legumes (Fabaceae) was observed, for example, *Vicia cracca*, *Vicia sativa* and *Lathyrus aphaca*. Compared to the control variant, abundant plants from the forage functional group grew in both burn variants (higher and lower intensity): *Foeniculum vulgare*, *Convolvulus arvensis*, and *C. cantabrica*, which were also the first to emerge three weeks after the 2019 induced burn. Total cover of renewed vegetation did not exceed 25% at either fire intensity after 24 months. Based on two years of research, we conclude that higher fire intensity does not allow survival of therophyte (monocotyledonous) life form, while hemicryptophytes and hamephytes (dicotyledonous) are more resistant to higher fire intensity and therefore have a greater possibility of survival.

Keywords: functional plant groups, hemicryptophytes, induced fire, therophytes, Mediterranean region

The link between growth crisis and soil cadmium contamination in maize

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Summary

Cadmium is a heavy metal present in vast array of ecological habitats including those intended for maize production. It is known that cadmium is detrimental for plant growth and health affecting photosynthetic performance. The aim of our study was to capture the effects of cadmium on maize photosynthetic performance during and after the growth crisis stage marking the transition of plant to autotrophic state. In this study, effects of different concentrations of cadmium on plant physiology and development in the early stages of maize plant growth were determined through the measurement of photosynthetic parameters as well as chlorophyll content. Two maize inbreds and their hybrid were grown under controlled conditions in a growth chamber and measured six times during four-week growth period in control (Co), 150 mg/kg (Cd150) and 375 mg/kg soil Cd (Cd375). Both Cd treatments significantly reduced plant biomass. Photosynthetic performance was not reduced until 14th day after sowing when the growth crisis is expected to occur, while from 15th to 28th day after sowing, steady decrease in photosynthetic performance was observed. Implications of these results will be discussed.

Keywords: cadmium, maize, soil, physiology

Validation of model for prediction the available amount of Mn in the soils of eastern Croatia

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Summary

The available concentrations of Mn in arable soils significantly depend on the pH value of the soil, the texture and the content of organic matter. Therefore, there is an assumption that it is possible to predict the available Mn fractions using the available analytical data on the soil. In previous research, models were created based on the analytical results (pH, SOM, soil texture, total and available Mn in the soil) of 229 soil samples. The aim of this research was to validate the model that predicts the availability of Mn in soils based on the analytical values of soil pH and SOM and for that purposes, 40 soil samples with a range of $\text{pH}_{\text{H}_2\text{O}}$ 5.47-8.61, pH_{KCl} 4.1-7.84, SOM 0.83-3.92 were analyzed. Also, plant available Mn extracted by EDTA solution (range 8.5-93.2 mg/kg) and total soil Mn extracted by aqua regia (range 145-759 mg/kg) were analysed. The range of predicted available Mn by validated model ($\text{Mn}_{\text{EDTA}} = 85.824 - 13.432 \times \text{pH}_{\text{H}_2\text{O}} + 6.044 \times \text{pH}_{\text{KCl}} + 2.2315 \times \text{SOM}$) was only 17.4-46.9 average error 16.3 mg/kg. Model predict insufficient availability of Mn in only 32.5% samples and according to real data 60 % of samples are in that category. Model accuracy could be significantly increased by including soil texture and total soil Mn data ($\text{Mn}_{\text{EDTA}} = 0,070 \text{ Mn}_{\text{AR}} + 5,205 \times \text{pH}_{\text{H}_2\text{O}} - 6,325 \times \text{pH}_{\text{KCl}} - 2,219 \times \text{SOM} + 5,5 \times \text{Text}$). The new model predicts 47.5% samples with insufficient Mn (should be 60%) and high available Mn in 22,5% samples (should be 27.5). Therefore, it can be concluded that the model for predicting the availability of Mn based only on basic soil properties is insufficiently precise, especially for soils poor or rich in available manganese, and that the accuracy of the prediction can be higher by including data on the content of total Mn and data describing soil texture.

Keywords: total Mn, soil texture, EDTA extraction, aqua regia, poor soil Mn

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The effect of physiologically active substances on the morphological properties of cherry fruit

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Summary

Cherry (*Prunus avium* L.) is an economically highly profitable fruit crop, due to the market, technological and commercial value of the fruit. The aim of this paper was to determine how treatment with different physiologically active substances affects individual morphological properties of the cherry fruit, Regina variety. Research of the effect physiologically active substances had on 75 trees in the cherry orchard was conducted in 2022, during the phenophase of fruit growth and development, in two locations in Zadar County: Ninski stanovi and Murvica. The treatment was carried out three times, with knapsack sprayers on five variants in 3 repetitions: K - no treatment, only water, T1 - CaO 15%, T2 - biostimulator (based on *Ascophyllum nodosum* L. extract), T3 - abscisic acid (ABA), T4 - salicylic acid. The first treatment was carried out at a certain time interval after full flowering, depending on the microclimate of the particular location, while the other treatments were conducted with intervals of 10 days. The fruit samples on which morphological measurements were performed contained twenty random fruits per repetition. Data processing with one-way analysis of variance (ANOVA) and regression test (Tukey) determined the effect of physiologically active substances on the length, width, weight, as well as on the percentage of dry matter and fruit toughness, by treatments and locations. The highest values of fruit length, width and mass were achieved by treatments T2 and T4, while the least cracked fruits were obtained with treatment T1.

Keywords: cherry, fruit, morphology, physiologically active substances, Zadar County



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Opportunities, challenges and limits in rural green tourism in Romania. A survey-based research

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Summary

The evolution of tourism in the last half century has been impressive, but not without turbulence, challenges and shocks. Within this evolution, the relationship between tourism, environment and communities gradually became an issue of major importance, and the increase in tourists' interest in nature, recreational and cultural activities in a clean environment, inherently generated serious concerns for preserving the quality of the environment, life, and local traditions. Implicitly, rural areas have become a favoured area for the manifestation of sustainable business-tourism models. On the one hand, a rural tourist destination relies on the quality of natural and cultural resources, but on the other hand the rural environment can also be influenced, both positively and negatively, by tourism activities. The increase of tourism activities in the global economy has transformed tourism in rural areas into a mechanism for the revival of rural communities and, in many cases, it is the subject of local or national public support. Starting from the natural attractions and diverse cultural contexts of the analyzed rural regions, this paper tries to identify the potential, opportunities, challenges and limits of rural green tourism in Romania. The research method is content analysis based on documents and in-depth interviews with several entrepreneurs in rural tourism, NGOs and other stakeholders involved in rural touristic activities. We investigate their concerns and perceptions regarding the impact of tourist activities on the environment, financial constraints, opportunities and limits of promotional activities, the importance of brand recognition and the image of the company associated with sustainable tourism, but also aspects related to personal responsibility, the authenticity of green efforts, and the correct orientation of public support towards sustainable tourism businesses in rural areas. We have also identified a series of challenges and difficulties that can block businesses from joining green rural tourism initiatives, such as the lack of knowledge and information about the concept of sustainable tourism, additional expenses, the lack of time and financial motivations necessary to integrate into these activities, the amount of additional work, the difficulty of obtaining certification etc. Not infrequently, the respondents suggest that integration into sustainable rural tourism is more difficult than they anticipated, and rewards such as increases in the number of customers, additional income, a favorable competitive position, recognition, seem still out of reach. Finally, we suggested a series of recommendations such as extensive promotion, accessible information, emphasizing the advantages in the medium and long term, the involvement of public and non-governmental organizations and campaigns to educate consumers about the importance of sustainability and the value of rural tourism.

Keywords: sustainable tourism, green practice, rural area, SMEs, Romania

The impact of perceived difficulties on the SMEs in the rural areas of Romania

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Summary

Small and Medium Enterprises are considered a major source of economic growth, employment, wealth creation and poverty alleviation in rural regions in both developed and developing countries. Understanding the characteristics and specific constraints facing the survival and growth of small businesses is essential in providing real and effective support for these firms, in line with how rural entrepreneurs perceive their needs and challenges. By applying a questionnaire and in-depth interviews with business owners, consultants, supporters and influencers in rural businesses in the North-West Development Region of Romania, we tried to compare and identify the differences in perception on the challenges of the rural space for small businesses. Specifically, we found that a large part of the expectations refers to the need to support local businesses, to reduce the prosperity gaps between regions and communities, especially between rural and urban areas. However, there are concerns, both among entrepreneurs and stakeholders, that unfocused political and economic interventions, not adapted to local constraints and particularities, can affect business growth, compromise opportunities and waste the few available resources. The preliminary results also indicate that the firms in the rural area are more concerned with changes in the economic and competitive environment, the accessibility of public support and skills shortages, and, to a lesser extent, perceive regulation and administrative burden as an obstacle for their survival and success. The study also reveals the importance of easy access to suitable financing, but also that the performance of SMEs has a bi-directional relationship with the accessibility of financing. Another important finding is that SMEs in rural areas significantly value the collaboration and exchange of information with companies from similar areas and contexts. In this sense, we advance the hypothesis that rural SMEs must develop collaboration as a tool for innovation and access to critical resources, in order to adapt to present-day conditions and challenges. Another significant observation is that, while aware of the importance of digitization, SMEs in rural areas perceive few short-term opportunities and advantages from the adoption of these technologies. Finally, we conclude that, beyond the need for geographically-differentiated support for small businesses in rural areas, it is the identification of some particularities that define businesses in the rural area that could better substantiate the economic policies in the field.

Keywords: rural areas, business perceived difficulties, constraints, opportunities, Romania, qualitative study

Agricultural policy development in EU pre-accession countries

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Summary

The paper presents the results of the study from the latest follow-up of the state of agricultural and rural development policy in the Western Balkan (WB) countries/territories and Türkiye and their approximation with the EU CAP, focusing on the years 2020 to 2021. To facilitate the analysis and comparison of the policy among pre-accession countries and the CAP, and following the EU two-pillar concept, this study uses the APMC tool as a uniform classification of the different policy measures among countries/territories. Different indicators derived from additional macroeconomic and sectoral data allowed comparison.

The pre-accession countries vary in the budget spent for agriculture and rural development. In the period 2020-2021, all WB countries increased the amount of total budgetary transfers to agriculture, whereas Türkiye has decreased it. The countries continued with almost the similar structure and trend in the budgetary transfers to agriculture and rural development as the previous period 2017-2019 (the first pillar dominating in all countries except Albania and Montenegro). The actual execution of agricultural policies remains with almost exclusively sectorial focus with the number one priority being increasing producers' income (support that is production-oriented), followed by the support for on-farm investments. Compared to the EU average, the total budgetary transfers presented per hectare and per inhabitant are still far below in all pre-accession countries.

Keywords: Agricultural policy, rural development, EU CAP, Western Balkan, Türkiye

Outlook on Croatian corn market using a partial equilibrium model

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Summary

Using the AGMEMOD (*Agricultural Member State Modelling*) sectoral partial equilibrium model a medium-term outlook of corn market in Croatia up to 2030 is simulated. We simulated future developments of sown areas, yields, production, imports, exports and corn prices under *ceteris paribus* market conditions in addition to the existing measures and instruments of the Common Agricultural Policy (*baseline*). The modelling results indicate the continuation of the positive corn market developments in Republic of the Croatia by the end of the simulated period. According to the *baseline* simulation results, the area sown with corn in Croatia will not change significantly, while the average corn yields will increase by over 10 t/ha until 2030. Considering the increase in yield, it is expected that production will also increase, while domestic consumption will not change significantly, which will result in a further increase in the net export of corn until the end of the simulated period.

Average producer prices are expected to begin to stabilize in 2024, but will still be higher on average compared to the observed historical period.

Keywords: medium term market outlook, AGMEMOD, corn, Republic of Croatia

Consumers' attitudes towards the origin of bakery products in Croatia

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Summary

Bakery products in Croatia as foods of high nutritional value, due to the busily lifestyle and price affordability, represent a basic foodstuff and an everyday part of the diet. According to the data from Croatian chamber of Economy, the average consumption of bakery products per inhabitant is 62 kg, which is 15.1% of the total expenditure on food. The aim of the research was to determine the opinions of consumers regarding the origin of bakery products and the influence of this information on the choice of buying these products. The research was conducted in 2021 on the territory of the Republic of Croatia using the CAWI quantitative research method on a representative sample of 1,022 adult respondents. The obtained research results confirmed a high level of purchase and consumption of bakery products, as well as a high level of awareness of the presence of semi-finished frozen mostly imported products on our market. Data analysis revealed that 70% of consumers are familiar with the country of origin of bakery products, and equally, 80% of them emphasized the importance of indicating the country of origin of the product. Further analysis indicates that 64% of the respondents do not base their knowledge about the origin of the product on the information on the product, but on the assumption that bakery products are always produced locally. A total of 92% of consumers believe that information about the origin of bakery products should be highlighted at the point of sale, and they associate the information about the origin with factors of product quality and freshness. In accordance with the results of the research, it can be concluded that when buying bread and bakery products, consumers have certain expectations that they associate with the quality and freshness of the product, and indirectly with the country of origin. This indicates that it is necessary to clearly and unambiguously inform consumers about the country of origin of bakery products, through recognizable labeling, as a confirmation of proof of traceability, quality and freshness of bakery products, but also awareness of the advantages of domestic products in the economic aspect, as a contribution to the sustainability and growth of domestic production.

Keywords: bakery products, country of origin, labelling, consumers' attitudes

The impact of Chinese business on the agricultural and food products market

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Summary

The purpose of this paper is to analyze the impact of Chinese business of agricultural products on the European Union market (import and export, the Covid-19 pandemic, and the African Swine Fever). Despite its strict policies, the country is open to foreign trade which represented 35% of its GDP in 2020. In recent decades, China has significantly opened up its market and has become the world's largest exporter and second-largest importer. The export of agricultural and food products in 2019 amounted to 64.83 billion dollars, which is an increase compared to 2005 by 85%. The part of global agricultural and food exports is 4.1%, and agricultural and food products are responsible for about 7% of the total export volume and 2.5% of its value. China's import of agricultural products increased by 13.9% compared to the previous year (\$171.94 billion in 2020). But, the appearance of African swine fever affected the decline of China's GDP, as also the increase in imports and the price of pork. Pork prices began rising in February 2019 and rose 154% over the next 11 months due to a severe supply shortage. Before the outbreak of fever, China was able to meet 97% of its pork consumption domestically, but supply shortages caused by swine fever quickly led to an urgent need to import from the global market. According to the General Customs Administration, compared to August 2018, pork imports increased by almost 102% in May 2019 and 314% in March 2020. Although China is one of the strongest economic countries, the research results showed that the emergence of African swine fever and the impact of the Covid-19 pandemic left a significant mark on foreign trade.

Keywords: China, agricultural product, foreign trade, Covid-19, African Swine Fever

Production Quotas, Competition and Farm Values

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Summary

This study provides an important contribution to the sparse literature on the effects of industry concentration (market power) on the value of Ricardian-type rents. The empirical stage is provided by the swine industry in North Carolina whose rapid expansion led the state officials to enact the moratorium on the construction of new production facilities to prevent further escalation of environmental problems associated with manure disposal. At the time of the imposition of the moratorium, the existing hog farmers were grand-fathered into the program by being issued free-of-charge production permits. Despite its original environmental policy objective, the moratorium effectively served as the barrier to entry into live hog production and significantly contributed to the rapid consolidation of downstream pork processors in the post-moratorium period. At the same time, the upstream live production segment, despite the quota program, remained competitive. Given the fact that future expansion of the live hog production was blocked, the production permits became the scarce factor and the source of future long-run profits (quasi rents). Excess profit earned by a farmer should be regarded as the true opportunity cost of that permit in the long-run. Under perfect competition, the equilibrium value of this rent would be determined by whatever it takes to drive profits to zero. Hence, the equilibrium price of live hogs would determine the rent, and not the other way around. These quasi rents would accrue to scarce factor owners (farmers) and not processors or integrators. The presence of the market power of processors on the market for live hogs (or the market for contract grower services) diminishes the value of that quasi rent below the perfectly competitive case. Consistent with economic theory, our empirical analysis shows a considerable decline in the value of the quota that is attributable to market concentration among processors. Because these permits are tied to specific properties and are not separately tradeable, we use a hedonic model to isolate the baseline permit value as one of the components of the bundle of property attributes. The interaction of the permit variable with various measures of buyer concentration enables us to estimate the change in the value of the permit as the concentration index changes over time. Using a digitized proprietary data set of farm sales in North Carolina from 1994 to 2010 we find that production quotas accounted for 31-50% of hog farm values. Farm profits from these production permits fell by 12-17% during our sample moratorium period, costing farmers between \$60,000 and \$68,000.

Keywords: agricultural policy, oligopsony, production quotas, hedonic models, land values.

Trends in Agricultural production in the Dubrovnik-Neretva County in the Pandemic years 2019-2021

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Summary

The specificity of the Dubrovnik-Neretva County is its exceptional seasonality of its population during the course of the tourist season. According to the 2021 census, 115,862 inhabitants were recorded, and during the record year in 2019, Dubrovnik-Neretva County has taken on 2,285,774 visitors. Therefore, there is a noticeable necessity for the vegetable and fruit crops as well as aromatic and spice herbs, within the production area of the county itself.

In the course of the past three pandemic years (2019-2021), the market relations changed significantly, and also have changed the requirements for agricultural products, and marketing and business conditions in the agricultural sector, respectively. The aim of this paper is to present the qualitative and quantitative trends in the exploitation of the agricultural land and the analyze the emergence of new ways of doing business during the pandemic years. For the purpose of the work, official statistical data on agricultural production of the Dubrovnik-Neretva County in the period of pandemic years (2019-2021) were used, which revealed certain trends that are caused by the above events during the specified period.

The ARKOD plots areas have increased from 9.625,85 ha to 9.898,37 ha, i.e., for 272,52 ha (2,83%). The largest increase was recorded in karst pastures (419,39 ha or 48,56 %), and for the moment at the unmaintained plots (for 44,99 ha or 60,43 %). The significant decrease was recorded in vineyards (88,37 ha or 4,16%) and orchards (50,05 ha or 2,11%).

The presence of fruit species of cultivated crops in the observed period has increased by 68,77 ha (what represents an increase of 3,06%). The largest part of the increase is referred to tangerines (119,56 ha) and for the mixed plantations of fruit species (34,01 ha), while the biggest decrease was recorded with the figs (44,61 ha).

For the vegetable species it was recorded an increase in cultivation area by 14,94 ha (or 3,86 %).

Preliminary data indicate a negative effect induced by the pandemic period on the smallest of agriculture economies in Dubrovnik-Neretva County and production that is demanding in terms of financial investment and consequently continuously high levels of maintenance and product placement (viticulture).

Keywords: Agricultural production, Dubrovnik-Neretva County, 2019-2021 Pandemic years, Quantitative indicators, Qualitative indicators.

The significance and economic role of Earth observation (EO) data in the agricultural development of the central Dalmatian islands

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Summary

In recent decades, climate change has been the focus of scientists, the media, and people in general. The impact of climate change is visible today, and to an even greater extent, the consequences are still expected in the future. The aim of this study was to reveal the inventory and applicability of innovative technological solutions in agriculture based on the usage of EO data dealing with climate change. The study analyzed the current agricultural situation, challenges, problems, and future trends in the development of agriculture in the central Dalmatian islands, with particular reference to the impact of climate change on island agriculture. The study inventoried and explained the possibilities of using EO data to cultivate Mediterranean crops on the four central Dalmatian islands from Split – Dalmatia County with a particular focus on the two most common agricultural crops - olives and grapes. In the end, the study resulted in the identification of implementation guidelines for the implementation of EO data in the cultivation of Mediterranean cultures in the research area. The use of products based on EO data can significantly improve the development of agriculture on the central Dalmatian islands, where the key prerequisite is systematic and high-quality education of agricultural producers, more active engagement of the holders of rural development in the public sector, and the implementation of tailor-made solutions for applying EO data in agriculture.

Keywords: Earth observation, agriculture, rural development, Dalmatian islands, Copernicus

Assessing the short-term impact of rural development grants in Croatia using a kvazi-experimental approach

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Summary

The objective of this research was to assess the impact of grants awarded under Operation 4.1.1 of the “Rural Development Program of the Republic of Croatia for the period 2014-2020” on the business indicators of the beneficiaries. A combination of Propensity Score Matching (PSM) and the Difference-in-Difference methods (DID) was used to estimate the average treatment effect on the treated (ATET). The analysis included data for 87 firms receiving grants (treated firms) and 12,804 other firms in the same sector (control group). We used 18 business indicators for each firm as covariates, and 13 as output variables.

The analysis covered the period 2018-2021, so the short-term ATET was determined. Grant receipt was found to have a positive effect on employment at recipients throughout the period ($p_{t+1} < 0.1$; $p_{t+2} < 0.05$; $p_{t+3} < 0.01$). A short-term positive effect was found on the debt ratio ($p_{t+1} < 0.1$; $p_{t+2} < 0.01$), and a less significant increase in average wage was recorded only in year $t+1$. For other indicators, the effect of treatment was not significant, i.e., the values were not significantly different from those of firms without support. We find similar results in the studies of other authors who evaluated the impact of grants from the EU rural development programs in the short term.

The conducted research shows the usefulness of using a counterfactual approach to evaluate the impact of public subsidies in agriculture, provided that the necessary data are available. The results obtained indicate a modest ATET, which could be a consequence of the narrow time span in this analysis. In any case, based on the analysis conducted, we recommend repeating this analysis with a longer time span, and introduction of regular assessments of public programs impact in Croatia using quasi-experimental methods.

Keywords: Croatia, rural development, propensity score matching, difference-in-difference, counterfactual evaluation



Session

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Genetics and Plant Breeding

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Efficiency of indirect selection for agronomic performance by multispectral vegetation indices

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Summary

Maize breeding can benefit from remote and proximal sensing in terms of understanding the biological complexity underlying the agronomic traits, as well as optimizing resource allocation in terms of indirect selection. Remotely captured vegetation indices (VIs) usually bear information on crop greenness, pigment/water content or chlorophyll fluorescence which is then analyzed in terms of usability in breeding programs. Fifty maize hybrids were screened in six experiments over two years and assessed for agronomic performance (grain yield and moisture). Multispectral screening was carried out by a six-band multispectral array (610, 680, 730, 760, 810, 860 nm) and fifteen normalized VIs were calculated. Sensor was set at 2m height, one meter within each plot and carried between plots during flowering. Partial least squares model was set with VIs as predictors and grain yield and moisture as response variables. Genetic correlations were calculated between predicted and observed agronomic performance and efficiency of indirect selection was calculated. Models explained 24 and 49% of variance for grain yield and moisture, showing high genetic correlations between predicted and observed values, resulting in 90% indirect selection efficacy. Our study demonstrated that remotely sensed VIs might be utilized for in-season performance predictions, reducing the costs of screening trials while increasing the information density.

Keywords: maize, indirect selection, multispectral, vegetation indices

This work is supported by the project “IoT-field: An Ecosystem of Networked Devices and Services for IoT Solutions Applied in Agriculture” co-financed by the European Union from the European Regional Development Fund within the Operational Programme Competitiveness and Cohesion 2014–2020 of the Republic of Croatia.

Assessment of phenotypic traits in lettuce under drought stress

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Summary

Phenotyping of genetic resources of lettuce focuses on growth and stress tolerance, but attention is also paid to the content of the bioactive compounds, such as flavonoids and anthocyanins. In our experiments, we tested the effects of growing conditions (greenhouse vs. outdoor) or drought stress on the contents of total flavonoids, anthocyanins and total phenolics in lettuce (*Lactuca sativa* L.). In parallel with the wet analyses, we have applied fast non-invasive measurements of fluorescence excitation ratio records and hyperspectral imaging in visible and near-infrared spectra. In drought stress variants, the PSII photochemistry was monitored using chlorophyll fluorescence imaging. The analyses have shown relatively tight correlations between the parameters derived from the fluorescence excitation ratio analysis and the flavonoid and anthocyanin content. In similar, the anthocyanin contents can be well estimated by the hyperspectral records. The quality-related growth and shape traits of lettuce were efficiently analyzed using the regularly captured RGB images. Drought stress led to an increase in flavonoid and anthocyanin content, but water shortage affected the growth. The fluorescence imaging analyses enabled the quantification of drought stress in individual genotypes. In summary, our results demonstrated the usefulness of high throughput non-invasive optical phenotyping tools to screen the lettuce germplasm according to the quality-related traits.

Keywords: lettuce, phenotyping, drought, non-invasive methods

The study was supported by grants VEGA 1-0664-22, and APVV-18-465.

Drought stress phenotyping of wheat genotypes using noninvasive methods

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Summary

Insufficient phenotypic data related to stress tolerance significantly limits the exploitation of genetic resources. Previously, we created a collection of 35 diverse winter wheat genotypes from Slovak Genebank at NAFC-RIPP Piestany, Slovakia, broadly covering a diversity of leaf traits. The phenotyping pot experiment was established with a whole collection of 35 wheat genotypes exposed to long-term (permanent) moderate water deficit compared to well-watered variant. The growth responses to drought were assessed by the automated phenotyping platform with image analyses. Moreover, the manually operated measurements of chlorophyll content (SPAD), chlorophyll fluorescence, leaf thermal imaging, and VNIR hyperspectral records were performed in 3-4 days intervals to assess the effect of drought stress on photosynthetic apparatus. The measurements were followed by analyses of aboveground biomass and grain yield. Our results indicated variations in responses to prolonged drought, such as decrease in plant height, an increase in canopy temperature, and decrease in leaf area index (LAI) due to the senescence of non-producing tillers. We observed a high diversity in the flag leaf area reduction, leaf chlorophyll content, spectral reflectance, and parameters derived from fast chlorophyll fluorescence. Moreover, we identified a close relationship between the changes in leaf traits and leaf optical properties measured by the hyperspectral reflectance records.

Keywords: wheat, phenotyping, drought, genotypes, noninvasive methods

The study was supported by the national grants VEGA 1-0664-22, and APVV-18-465.

Quality control of insecticide-treated corn seed using the Heubach test

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Summary

The treatment of corn seeds with some of the phyto pharmaceutical preparations is a standard measure of seed protection, and the sowing of such seeds ensures the protection of the young corn plant from diseases and pests. Seed treatment must be in accordance with environmental protection and a high degree of care for the health of living organisms. Measuring the dedusting of fine dust particles from the seed surface is a reliable method for investigating the quality characteristics of seed treatment and is performed by the Heubach test.

The average amount of particle dedusting in two years was 1.0349 g/100 kg of treated seeds. The research of the quality characteristics of the treatment showed that the highest values of fine particle dedusting were recorded in the coarse fraction of seeds in both years of the research, which was 1.2661 g/100 kg on a two-year average. The measured values of particle dedusting in the fine and medium seed fractions are lower and on a two-year average they are 0.9406 g/100 kg in the fine fraction and 0.918 g/100 kg of seeds in the medium fraction. The results of the research undoubtedly indicate that the treatment was performed correctly, and that by sowing properly treated seeds of corn hybrids, there are no risks of impact on the environment and human health. The aim of the research is to analyze the quality characteristics of seed treatment of corn hybrids treated in two production years.

Keywords: corn, seed treatment, Heubach test, environment, sustainability

Limited transpiration and Liebig's Law of minimum in hot, managed environments

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Summary

Increasing vapor pressure deficit (VPD) due to climate change has adverse effects on maize yields. Increasing VPD implies increased evapotranspirative demand which can't be sustained in most of the rainfed maize growing regions limiting the water use efficiency and thus grain yield. Recently, it was found that the differences in stomatal conductance between maize genotypes in high-VPD conditions can be attributed to variability in hydraulic conductance of conductive tissues, thus limiting transpiration. We carried out multi environment trials (MET) over five consecutive years (2015-2019) in 18 managed environments in Turkey with 141 experimental hybrids. All environments were hot with mean temperatures in July and August higher than 25°C. However, in some environments (Adana, Mersin), high mean relative humidity resulted in decreased VPD. Mean annual grain yields were 16.94, 14.4, 15.7, 15.2 and 13.44 t/ha. It was found that genetic variance linearly increases with increasing VPD with coefficient of determination of 99%. Genotypes were divided into two groups the ones above the genotype by environment (GEI) median BLUP (A_{med}) and the ones below the GEI BLUP (B_{med}). Simple linear regression models showed linear increase of grain yields with increasing VPD in A_{med} group ($R=0.54$) and decrease in B_{med} group ($R=0.52$) corroborating our previous findings. The results of our study will be discussed considering Liebig's Law of minimum and yield potentials approaching the water and VPD ceilings.

Keywords: maize, vapor pressure deficit, yield, managed environments

Selection of winter hardiness *Pisum sativum* L. genotypes based on phenotypic characterization

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Summary

Climate change is currently one of the biggest challenges in the breeding process and crop production. Pea varieties are usually sown in the spring, and the yield they produce depends greatly on the temperature and water availability during the flowering phase. Autumn-sown varieties have a higher and more stable yield than spring-sown varieties. They usually enter the flowering phase earlier to avoid stresses due to high temperatures and drought, which are frequent in spring. Overwintering is a complex quantitative trait influenced by the genotype and the environment in which the plant is grown. Phenotypic evaluation is the first step in identifying potential resources in breeding programs for creating an initial breeding population. The aim of this study was to evaluate the overwintering of dry pea accessions in the field. The degree of freezing tolerance for all pea varieties was investigated under field conditions over a period of three years. Additionally, the degree of regeneration as well as the sum of the blossoming temperature were estimated. Convar. *sativum* has been found to be more cold tolerant than convar. *medullare*. Late maturing accessions had significantly better tolerance to winter conditions. Based on the obtained data, nine accessions were selected because of their strong cold tolerance. Their hybridization is expected to produce an initial population for the development of a new breeding program of dry pea.

Keywords: *Pisum sativum* L., winter hardiness, accessions, field trials

The influence of drought on morphological, physiological and biochemical characteristics of two ecotypes of garlic

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Summary

Drought negatively affects plants by altering morphological, physiological, and metabolic processes. Therefore, yield reductions under drought conditions are the result of plants being affected by drought. Plants have developed various mechanisms to defend themselves against water deficiency. Garlic is an important member of the Alliaceae family and plays a significant role in agricultural production. Garlic is sensitive to water limitation and drought stress, and maximizing the yield of garlic bulbs depends mainly on water availability. The objective of this work was to determine the effects of drought on morphological and physiological traits, as well as on amino acids and sugar content in two ecotypes of Istrian garlic (Istrian red and Istrian white). Drought was induced in garlic plants by using PEG solution. The osmotic potential of PEG solution was - 0.60 MPa. The multispectral measurements were performed using CropReporter™. Drought stress affected plant weight, plant length, number of leaves, chlorophyll index, maximum quantum yield of PSII, maximum chlorophyll fluorescence, and inulin and glucose content in both ecotypes. On the other hand, an increase was observed in root length, red reflectance, free amino acids content and sucrose content under dry conditions. Based on the obtained results, it was determined that Istrian red is more tolerant to drought stress than Istrian white ecotype.

Keywords: garlic, drought, morphological traits, multispectral analysis, amino acids

Micropropagation and microtuberization of the traditional potato cultivar 'Brinjak'

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Summary

The traditional potato cultivar 'Brinjak' has been grown in Lika (a region in Croatia) for many generations, and the producers' interest in its revitalization is great. The cultivar is resistant to diseases caused by the pathogens *Phytophthora infestans* and *Streptomyces scabies*, but it is infected with potato viruses M and S, which reduce yield. To bring out the full genetic potential of the cultivar, the initial generations of seed potatoes need to be propagated from healthy mother plants by micropropagation or microtuberization. The aim of this work was to examine the success of micropropagation and microtuberization in the potato cultivar 'Brinjak' under different treatments. MS medium with the addition of 3% sucrose was used for micropropagation, and medium with 8% sucrose for microtuberization. Plants micropropagated in medium without growth regulator (HFM) and medium supplemented with 2.5 mg/l GA3 had the best shoot quality, root development and rooting percentage. In the case of microtuberization, a higher percentage of shoots with formed microtubers was obtained in the treatment with 4 mg/l kinetin and 1 mg/l 6-benzylaminopurine (KIN4 BAP1) compared to the treatment without growth regulator (HFM). In addition, darkness had a favorable effect on a higher percentage of shoots with formed microtubers. The diameter and average microtuber weight were greater in the KIN4 BAP1 treatment under short days. In conclusion, both methods can be successfully used in the propagation of healthy mother plants, but with micropropagation it is possible to obtain many more plants in the same period of time.

Keywords: *Solanum tuberosum* (L.), micropropagation, microtuberization, plant growth regulators, photoperiod

Assessment of agronomic values and stability of yield components of newly created spring field pea lines (*Pisum sativum* L.)

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Summary

Field pea (*Pisum sativum* L.) is one of the most important protein legumes. Although grain yield is the most important breeding trait, the focus of the breeding process is also directed at other quantitative traits (yield components). The study of genotypes (breeding lines) in different environments through the analysis of the most significant quantitative properties, as well as the assessment of their interaction values, contributes to the creation of new sources of genetic variability and new genetic material (varieties/hybrids). The study goals were to evaluate the agronomic value of nine newly created spring breeding lines of fodder pea during 2019 and 2020, to evaluate the stability of yield components and to select the most promising genotypes for further breeding procedures based on the obtained results. The results of the study showed that there are significant differences between the tested genotypes in the tested environments for most of the analysed properties and that the variation of genotypes has the greatest influence within the identified sources of experimental variation. The stability assessment was made by the grouping method according to Francis and Kannenberg, based on which the values of the genotypes L – OSMBK 1, L – OSMBK 3 for grain/plant yield L – OSMBK 1, L – OSMBK 8, L – OSMBK 3, L – OSMBK 9 for 1000 seed weight and L - OSMBK 9 for the plant height were in group I, which is characterized by high values and a small coefficient of variation of properties and represents the most stable and most promising breeding genotypes.

Keywords: spring field pea, yield components, stability, breeding material

Distribution of Rht, Ppd and Vrn alleles in diverse wheat germplasm

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Summary

The aim of this study was to determine the distribution of Rht (reduced height), Ppd (photoperiod) and Vrn (vernalization) alleles in diverse set of wheat genotypes. The study was conducted on 120 hexaploid wheat varieties from 24 countries, released between 1905 and 2010. Genomic DNA was extracted from fresh leaf tissue using CTAB (cetyltrimethylammonium bromide) method. PCR analysis was performed using functional markers specific for Rht-B1, Rht-D1, Ppd-B1, Ppd-D1, Vrn-A1, Vrn-B1, Vrn-B3 and Vrn-D1 loci. The presence of Rht-B1a allele (wild type) was determined in 47 % of varieties, and Rht-B1b allele (dwarf type) in 45 % of examined varieties. At Rht-D1 locus, most of the examined varieties (84 %) had the wild type allele Rht-D1a. Regarding the Ppd-B1 locus, most of the examined varieties (97 %) had Ppd-B1b allele (photoperiod sensitive), while on the Ppd-D1 locus 63 % of varieties had Ppd-D1a allele (photoperiod insensitive). At the Vrn loci recessive alleles vrn-A1, vrn-B1, vrn-B3 and vrn-D1 prevailed with frequencies of 88 %, 92 %, 96 % and 94 % respectively. It was observed that the distribution of alleles at tested loci differed based on the year of release and geographic origin of the examined wheat varieties.

Keywords: wheat, functional markers, Rht, Ppd, Vrn

Stability of yield and grain quality of winter wheat under different nitrogen fertilization levels

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Summary

This study was conducted to determine the stability of grain yield (GY) and grain protein content (GPC) in 25 winter wheat genotypes grown in 16 environments (combinations of two locations, two growing seasons, and four nitrogen fertilization levels). The AMMI analysis revealed significant effects of genotype (G), environment (E), and GE interaction for GY and GPC. Environment explained 74% and 76% of the treatment sum of squares for GY and GPC, respectively, while G and the GE interaction accounted for 11% and 15% of the treatment sum of squares for GY and 20% and 4% of the treatment sum of squares for GPC, respectively. AMMI 2 biplot revealed various levels of stability of the best-ranked genotypes for GY and GPC. The Finlay and Wilkinson regression coefficient (b) showed a moderate positive correlation ($r=0.40^*$) with mean GY, indicating that the best-ranking genotypes were better adapted to higher-yielding environments. In contrast, the correlation between b and mean GPC was not significant, suggesting that genotypes are not specifically adapted to particular environments and that simultaneous selection for mean performance and stability of GPC would be possible. This was also confirmed by the moderate negative correlation between mean GPC and the coefficient of variation (-0.50^*), suggesting that the GPC of the best-performing genotypes is less affected by different environments.

Keywords: wheat, yield, quality, AMMI analysis, stability

PHS differences in Croatian wheat cultivars

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Summary

Pre-harvest germination (PHS) is the premature germination of seeds at which we can see germination of germs on unharvested ears in the field. Some of the visible signs of PHS are seed swelling, sprout discoloration, separation of the seed coat from the core and the formation of roots and shoots. The intensity of PHS mostly depends on weather conditions, therefore there are variations from year to year. The aim of this research was to determine the influence of weather conditions on 10 wheat cultivars during two vegetation years (2017/2018 and 2018/2019). The research was conducted in two repetitions for each examined year. In the harvest, five ears were selected for each repetition. The ears were placed in containers with sand at room temperature and watered for 14 days. After a certain amount of time, each spike was rated on a scale of 1-7 (1 – no germination, 7 – whole spike germinated). A strong influence of the weather conditions on the research results was determined. In the first vegetation year, twice the amount of precipitation (July) was determined compared to the multi-year average, and significantly higher grades were noticed for each individual ear in comparison to the second year of testing, whose ear values were significantly lower.

Keywords: PHS, weather conditions, wheat cultivars

Variation of fusarium ear rot and mycotoxin concentration in grains of maize hybrids

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Summary

One of the most economically important pathogens of maize is *Fusarium verticillioides* (Saccardo) Nirenberg which causes Fusarium ear rot of maize (*Zea mays* L.). In addition to its effect on yield and seed quality, *F. verticillioides* can produce several toxic metabolites, of which fumonisins are most prevalent. In a three-year study, the resistance of 21 maize test hybrids to Fusarium ear rot was evaluated under conditions of natural infection and artificial infection after ear inoculation with the fungus *F. verticillioides*. The severity of the infection was estimated by visual score of ear infection, while the level of fumonisins B1, B2, B3 and B4 in the grain was determined by HPLC-MS/MS method. Combined ANOVA showed a significant effect of year and genotype as well as a significant genotype × year interaction for all examined traits, while the interactions of year × infection condition and genotype × infection condition were only significant for fumonisin content. The phenotypic correlations of the visual score of ear rot and individual fumonisins ranged from 0.69 to 0.77 under natural infection and from 0.60 to 0.65 under artificial infection. The higher heritability of the visual score as well as grain fumonisin content under conditions of artificial infection indicates the need to perform artificial inoculation for a more accurate assessment of genotype resistance to Fusarium ear rot in maize.

Keywords: maize, Fusarium ear rot, artificial infection, fumonisins

Potential of different genomic selection models for predicting wheat dough rheological properties

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Summary

When breeding for wheat quality traditional methods can be time-consuming and imprecise since the majority of quality traits exhibit complex inheritance and interaction patterns. Therefore, methods based on molecular markers, such as genomic selection, are getting increasingly used in wheat breeding. We examined the potential of five different genomic selection models for predicting five wheat dough quality traits – wet gluten content (WGC) and mixograph traits: optimal dough development time (MPT), dough consistency and stability at the end of mixing (MTW), energy used during the mixing (MTI), and dough strength (MPH). The population comprised of 153 recombinant inbred lines and the experiment was carried out for three years at two locations in Croatia. Genomic predictions were done on the basis of 2231 SNP markers. The highest mean prediction accuracies (r_{PA}) were observed for MPT (0.48–0.57), followed by MTW (0.48–0.54), WGC (0.32–0.47), and MPH (0.29–0.42). The lowest r_{PA} were obtained for MTI suggesting that this trait is not an ideal trait for genomic selection. The highest mean prediction accuracies for MPH and MTI were obtained using BayesLASSO model; for MTW and MPT using Random Forest model; and for WGC when using the Reproducing Kernels Hilbert Spaces model. All traits had high broad-sense heritabilities ($H^2 \geq 0.72$). The results showed that genomic selection may be an effective tool for predicting dough quality traits and selecting the best performing lines.

Keywords: wheat, dough quality, mixograph, genomic selection, prediction models

Changes in allele frequencies and genetic diversity in red clover after selection for cold tolerance using SSR marker

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Summary

The selection of populations of red clover (*Trifolium pratense* L.) adapted to extreme environmental conditions is of great importance due to constant climate change. The plant material analysed with SSR molecular markers included two parent populations (cultivars) and their reselections (breeding populations), which were created after one cycle of selection under freezing temperature conditions. Reselections PS1 and PS3 were produced by intercrossing 38 surviving plants of parent populations P1 (cultivar 'Reichersberger') and P3 (cultivar 'Croatia'), respectively. Total of 48 genotypes from each cultivar and each reselection were randomly selected for SSR analysis. 16 SSR primers were selected taking into account their presence of loci on all seven red clover chromosomes. A slight increase in the average frequency of alleles from the initial populations to the populations after one cycle of selection was observed, followed by a decrease in the number of alleles. Out of a total of 16 loci, Waples neutrality test revealed significant frequency changes at 12 loci in P1 to PS1 and 9 loci in P3 to PS3 populations. At these loci, the change in allele frequency is greater than expected due to the action of genetic drift alone, so these loci can be considered selectively non-neutral. The genetic diversity in the studied populations did not change significantly after selection, leaving enough genetic variability as a prerequisite for the success of future selection.

Keywords: *Trifolium pratense* L., abiotic stress, allele frequencies, microsatellites

Green mass and seed yield potential of alfalfa breeding populations

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Summary

Alfalfa is the most important forage leguminous in the production of voluminous fodder. It is not primarily produced for the seeds, but for the recognition and commercial viability of the cultivars on the market, in addition to the green mass yield, the seed yield is also important. Creating a cultivar of superior yield and forage quality with satisfactory seed production is one of the biggest challenges for alfalfa breeders and seed producers. The objective of the work was a during a five-year research (2014-2018) to determine the yields of green mass and seed of 19 newly developed alfalfa populations (ABP 1-19) of the Agricultural Institute Osijek and to choose superior materials for our breeding work and/or application of a potential future cultivar. Statistically significant differences were found between the studied ABPs in the average three-year (2014-2016) yield of green mass, which ranged from 68.41 t/ha (ABP 6) to 78.05 t/ha (ABP 19) and the average two-year (2017-2018) seed yield that varied from 150.78 kg/ha (ABP 9) to 335.35 kg/ha (ABP 7). The highest average annual yield of green mass (90.24 t/ha) was achieved in 2015, and the seed yield (394.17 kg/ha) in 2017. Several alfalfa populations (ABP 19, 8, 14) superior in both analysed traits were identified for the application of a potential future cultivar. Also were identified populations with high yield of green mass (ABP 12, 18) and seed yield (ABP 7, 4) which represent valuable genetic material for our further breeding work.

Keywords: green mass yield, seed yield, populations, alfalfa, breeding

Interrelationships of vegetation length, plant height and grain yield in winter wheat VCU experiments

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Summary

As part of the registration of varieties of agricultural plants, official variety trials are set up to test the value for cultivation and use (VCU trials). In accordance with the Ordinance on the registration of varieties of agricultural plants (Official Gazette 99/08, 100/09, 109/10, 77/13 and 57/18), the following data are determined for wheat varieties during the growing period, among others: sowing and emergence dates, plant height (at four sites), date of ear emergence (at two sites) and date of harvesting. At harvest, the following are determined for each plot: grain yield, hectolitre weight and grain moisture content. Data were used in the study from VCU wheat trials in the growing period 2019/2020, 2020/2021 and 2021/2022 at the locations Tovarnik, Osijek, Kutjevo and Zagreb. The trials were arranged in a randomized complete block design with four replications. Objective of this study was to examine the interrelationships of vegetation length, plant height and grain yield in VCU experiments of winter wheat and to express it with correlation coefficients. There is a statistically significant correlation between plant height and grain yield at the location Tovarnik in 2021/2022 and at the location Kutjevo in 2019/2020. There is a statistically significant correlation between the length of vegetation and grain yield at the location Zagreb in 2020/2021.

Keywords: VCU trials, winter wheat, vegetation length, plant height, grain yield, interrelationship

Changes in allele frequencies after four cycles of recurrent selection in the Maksimir 3 synthetic maize population

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Summary

The effects of four cycles of recurrent selection on simple sequence repeat (SSR) marker allele frequencies and population structure were examined in the Maksimir 3 Synthetic (M3S) maize population (*Zea mays* L.). Genotyping of 32 plants from each cycle of selection at 38 SSR loci revealed that the mean number of alleles per locus and average gene diversity were preserved over cycles of selection, indicating maintenance of sufficient genetic variability in the population needed for future genetic gain. Waples' test of selective neutrality revealed that genetic drift was the main force in changing allele frequencies in the population. The proportion of selectively nonneutral loci in single cycles of selection varied between 16% and 37%. Some of nonneutral loci shared the same genomic locations with previously published QTLs controlling important agronomic traits. Between 5% and 29% of loci were found to be in significant Hardy-Weinberg (HW) disequilibrium with majority of them showing an excess of homozygosity. Excess of homozygosity at several loci was highly consistent across cycle populations suggesting positive assortative mating as the possible cause of the observed HW disequilibrium. Linkage disequilibrium (LD) test revealed that the M3S population was essentially in linkage equilibrium. The proportion of pairs of loci in significant LD varied across cycles of selection between 0.1% and 1.8% probably due to the effects of genetic drift and epistatic selection.

Keywords: maize, selection, microsatellites, population structure



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Cryptogams in dynamics of sandy grasslands: specific response to grazing enclosure

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Summary

Cryptogams (lichens, mosses), though often neglected in the studies on vegetation dynamics, compose a large part of biomass and largely contribute to the biodiversity in sandy grasslands. We aimed to 1) explore the response of cryptogams to management changes and 2) contribute to the knowledge of dynamics of cryptogams in dry grasslands. Study objects were permanent plots in endangered sandy grasslands of ‘Pannonic inland dunes’ (EU Habitat Directive, 2340). Vegetation dynamics of *Corynephorum canescentis* (CC) and that of *Festucetum vaginatae* (FV), the two characteristic vegetation types of the Nyírség’s sand region, have been monitored in grazed and experimentally fenced stands. Fencing was used to model the prevailing trend of falling stocking densities. Biomass of cryptogams has been sampled in 2013 and 2018 (five and ten years after grazing enclosure, respectively), then species were separated under stereo microscope. Spot tests and HPTLC have been applied to identify critical lichen taxa. Fencing has led to an increased biomass of cryptogams within a few years. Lichens in general benefited relatively more from enclosure than the mosses. The increase of lichen biomass (especially of *Cladonia rangiformis*) in both communities is clearly due to the 10-year absence of grazing. The only lichen favoured moderate grazing has been the legally protected *C. magyarica*. Short spells of low intensity grazing can promote species-richness of cryptogams in the studied dry grasslands.

Keywords: grazing, lichens, mosses, *Corynephorum canescentis*, *Festucetum vaginatae*

The application of the EMBAL methodology to assess grasslands of high nature value

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Summary

During the period from 2020 to 2022, assessment of effects on high nature value grasslands was carried out for the implementation of two activities (10.1.3 Preservation of high nature value grasslands and 10.1.5 Pilot measure for the protection of butterflies). The survey of grassland vegetation was done using the standard Braun-Blanquet method, while additional survey was carried out using the methodology based on EMBAL (*European Monitoring of Biodiversity in Agricultural Landscapes*). The research included mapping and analysis of land cover according to the EMBAL classification in an area of 250 m width around 90 transects, distributed within three biogeographical regions in Croatia, where the butterfly survey was conducted. Within each mapped area, the vegetation was surveyed on at least 3 transects (2.5 m x 20 m), where data on composition and abundance of plant species was collected along with data on pollinator habitat suitability parameters (variety of flowering species colours, flower density, vegetation vigour, ratio coverage of graminoids in relation to forbs). Preliminary comparison of vegetation data and surveyed parameters does not show a significant difference between the transects depending on whether they are managed or not under the abovementioned activities. The analysis of the data collected by the Braun-Blanquet method will show whether the EMBAL methodology can complement standard methods in the assessment of high nature value grasslands.

Keywords: biodiversity, pollinators, vegetation, monitoring

Conservation and sustainable use of genetic resources of medicinal and aromatic plants in Croatia

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Summary

Plant biodiversity is the basis for sustainable agricultural production. Due to various geographical, climatic and soil conditions, Croatia has a great richness of medicinal and aromatic plants (MAP), whose natural populations show great diversity at the morphological, biochemical and genetic levels. However, they are threatened by genetic erosion caused by various factors such as habitat loss and fragmentation, as well as overexploitation. Within the framework of the National Program for Conservation and Sustainable Use of Plant Genetic Resources for Food and Agriculture in the Republic of Croatia and the Working Group for Medicinal and Aromatic Plants, MAP genetic resources are collected, conserved, characterized, evaluated, regenerated, and documented. The collected accessions are stored in *ex situ* conservation facilities at +4°C. Currently, the collection consists of 2652 accessions of more than 230 plant species. To fill the existing gaps in the MAP collection, collection missions are organized every year. These missions use the standardized collection form, which includes taxonomic information, taxon biology, a description of the collection site and vegetation, environmental data, an assessment of population diversity, and information on the risk of genetic erosion. The characterization and evaluation of the collected accessions is based on standardized, internationally accepted descriptor lists. After characterization and evaluation, the collected material is available for introduction into agricultural production and for future scientific studies and breeding programs.

Keywords: accessions, Croatian Plant Genetic Resources Database, diversity, ecogeographical survey, passport data

Flora of the Brijuni National Park

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Summary

The flora of the Brijuni National Park has not been systematically researched for years, and Makowsky conducted the last such research in 1908. Recently, the flora of Mali Brijun was investigated by Bogdanović and Ljubičić in 2018, while the flora of Veli Brijun and 12 neighbouring islands and islets remained completely unexplored. Field research was conducted during 2020-2022 in order to floristically investigate and to map the vascular flora of the entire Brijuni NP. A total of 759 taxa were identified, of them 202 for the first time by this research. There are total of 102 families, of which the most represented are grasses (Poaceae) with 86 taxa and legumes (Fabaceae) with 85 taxa. Out of the total number, 30 species are identified as endangered. Only one endemic species (*Limonium cancellatum*) was recorded in the flora. Regarding the life forms in the flora of Brijuni NP, the most numerous are therophytes with 307 taxa, hemicryptophytes with 233 taxa, phanerophytes with 78 taxa, geophytes with 71 taxa, hamephytes with 58 taxa and hydrophytes with 11 taxa. Cultivated plants are represented with 74 species. There are 14 invasive species and one new potentially invasive species (*Nassella neesiana*) for the Croatian flora.

Keywords: Istria, flora mapping, biodiversity conservation

Synonymy and homonymy – main problems in conservation of olive genetic resources

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Summary

The estimated number of olive cultivars in the world is 2600. Only a few cultivars have gained the reputation of worldwide importance, such as Arbequina or Koroneiki. Most olive cultivars are kept in collections with the aim to identify, protect and evaluate the genetic resources of the species. There are 2098 accessions listed in the Genesys database, originating from Spain, Italy, Croatia, etc. The largest network of olive gene banks is organized by the International Olive Council (IOC) and includes 23 germplasm and three international collections in Spain, Morocco and Turkey. The main problem in managing the collections is olive cultivar mislabeling during field expeditions as the result of frequent cases of synonymy and homonymy. Field expeditions require considerable background knowledge on the cultivar phenological traits, possible environment - cultivar interactions and expression, but also on the history of migrations (cultivar introduction) and traditional agricultural practices (propagation methods). Older and wider spread cultivars usually have a larger number of denominations (synonyms), such as our main cultivar Oblica (Croat. oblo = rounded) with more than 20 different synonyms. Very often the names of olive cultivars are linked to the most specific phenotypic characteristics and/or geographical origin. In fact, many different cultivars bear very similar or identical names (cases of homonymy) based on fruit colour (Bjelica, Belica [Croat. bijelo, dial. belo = white]) or origin (Divlja, Divikuja [Croat. divlje, dial. divije = wild]).

Keywords: gene bank, collection, management, cultivars, Olea

Identification of firefly species in Croatia and their morphological characteristics

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Summary

Fireflies (Coleoptera: Lampyridae) are among the most charismatic of all insects. With more than 2000 species in 100 genera, the worldwide diversity of lampyrids is impressive and includes both diurnal and nocturnal species. Fireflies, like numerous other insects, have experienced a worldwide decline in their distribution and abundance. Currently, 64 species of fireflies are known in continental Europe. In Croatia, seven species have been described and voucher specimens are kept in natural museums in Zagreb, Split and Varaždin. The last revision of fireflies in Croatia was made by Mikšić in 1986, who stated that the common firefly, *Lampyrus noctiluca* (Linnaeus, 1767), *L. zenkeri* Germar, 1817 and *L. lusitanica* Motschulsky, 1854 occur in Croatia. *L. noctiluca* is the most common of the three species and is widespread on the Croatian mainland, while it is rare in coastal areas. After a detailed analysis of the evidence from the Citizen Science Project “Krešo Krijesnica”, the following four species of fireflies were identified in the Republic of Croatia: *L. noctiluca*, *Lampyrus germariensis* (Jacquelin du Val, 1860), *Lamprohiza splendidula* (Linnaeus, 1767), *Luciola italica* (Linnaeus, 1767). Identification was based on the color of the emitting light and the appearance of the luminescent organ. The most important morphological characteristics and identification keys will be presented at the symposium.

Keywords: lampyridae, citizen science, luminescence, light pattern

Native vascular flora of special forest vegetation reserve of Kočje (island of Korčula)

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Summary

Kočje was protected as a special forest vegetation reserve in 1962. It is located on the island of Korčula and occupies an area of 4.62 ha. The Kočje area, which has preserved the appearance of untouched nature, is of geological, botanical and aesthetic value. In view of the successive changes in the vegetation and the fact that no systematic botanical research has been carried out in the last decades, the aim of this study was a preliminary inventory of the plant taxa of the area. Field research was conducted in late 2021 and the first half of 2022. A total of 125 plant taxa (123 species and two subspecies) were found. The taxa belonged to 40 families. Part of them (19) have now been registered for the first time. The most dominant families are: *Fabaceae* (13%), *Lamiaceae* (8%), *Poaceae* (7%), *Asteraceae* and *Liliaceae*. Phytogeographical analysis showed that the Mediterranean floral element is the most represented. The equal representation of Hemicryptophytes (30%) and Therophytes (27%) indicates a great influence of the Eumediterranean zone. Five endemic and 14 endangered species were identified, among which the *Campanula portenschlagiana*, which is both endemic and near threatened (NT), should be highlighted. Such a large share (11.2%) of endangered taxa gives this area a special significance and importance.

Keywords: flora inventory, biodiversity conservation, Kočje, Korčula

Chemical diversity of essential oil from lavandin (*Lavandula × intermedia* Emeric ex Loisel) landraces in Croatia

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Summary

The cultivation of lavandin (*Lavandula × intermedia* Emeric ex Loisel) has a long tradition in Croatia. Cultivation began on the islands of Hvar and Vis, from where it spread to the mainland. The high yield and good quality of the essential oil enabled the cultivation to maintain through the years. The aim of this work was to thoroughly investigate the essential oil composition of landraces originating from two islands (Hvar and Vis) and one from the mainland (Jastrebarsko). A total of 18 samples were analyzed by gas chromatography-mass spectrometry to determine their chemical profile. The main compounds determined were linalool, 1,8-cineole, linalyl acetate and camphor. The results of the study showed differences between landraces in the content of certain compounds. The study deals more closely with the differences in the content of camphor and linalyl acetate, which are often used as parameters for determining the quality and, at the same time, the price of the essential oil. By chemoprofiling the compounds, the best specimens can be selected for use in aromatherapy and cosmetics.

Keywords: lavandin, *Lavandula × intermedia*, essential oil composition, GC-MS

Plant diversity and plant invasions across different habitats of Murter Island (Croatia)

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Summary

The spread of alien species has been changing the diversity of plant communities in all parts of Europe and the world. Murter is an inhabited island rich with diverse habitat types and is part of the NATURA 2000 network. We conducted an analysis of 269 phytosociological relevés to detect differences in plant diversity and degree of invasion among habitat types on Murter island. Relevés were classified into different EUNIS habitat types using the expert system. The effects of local environment on the species α - and β -diversity of analyzed plant communities were also investigated. Alien species were present in 25% of the relevés and represented 5% of the total number of species. The habitats with the highest alien species richness were arable lands and gardens followed by Mediterranean subnitrophilous annual grasslands. The analysis emphasized that the habitats with a higher degree of invasion were the most disturbed ones and that in general habitats rich in native species did not harbor less invaders than habitats poor in native species.

Keywords: alien species, plant communities, diversity, NATURA 2000, habitat

Genetic tools and approaches for conservation management of *Mauremys rivulata* in Croatia

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Summary

The Balkan terrapin (*Mauremys rivulata*) is an endangered species protected by the Habitats Directive. Its range in Croatia is restricted to the southernmost karstic aquatic habitats. Most of these habitats are anthropogenic modifications of lentic and lotic water used historically for agriculture. The aim of this study was to investigate the available genetic tools for conservation management of the species and to conduct a preliminary analysis of the genetic diversity of *M. rivulata* in Croatia. The survey of literature and openly accessible genetic repositories revealed the knowledge on complete and incomplete genes, microsatellite markers, rRNA and complete mtDNA for the species. Amplification of all 18 microsatellite markers available for the species on 56 samples resulted in 15 successfully amplified markers. Nine of them were estimated to have high probability of null alleles and were excluded. Preliminary genetic diversity analysis of two populations from Croatia (Majkovi and Konavle) and one from Montenegro (Sutorina) based on six microsatellite markers revealed a high level of polymorphism (24 alleles per locus on average). The values of expected heterozygosity ranged from 0.85 (Sutorina) to 0.93 (both Majkovi and Konavle populations). Bayesian analysis of population structure revealed the existence of three gene pools, the first dominant in the Sutorina population, and the second in both Croatian populations, while the third was present to a lesser extent in all populations. Although they provide insights into the population genetics of *M. rivulata*, the available microsatellite markers are limited due to the high proportion of null alleles, especially for parentage analysis. SNP markers developed using the ddRAD method would provide a more precise and cost-effective estimate of effective population size and efficient population management of this threatened species.

Keywords: genomic SSRs, genetic diversity, population structure, *Mauremys*



Session

5.1 Book of Abstracts

Vegetable Growing, Ornamental, Medicinal and
Aromatic Plants

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Povrčarstvo, ukrasno, ljekovito i aromatično bilje

Selected parameters of carrot juice depending on the foliar application of selenium

Ivana Mezeyová, Alžbeta Hegedúsová, Tatiana Jaloviarová, Oliver Firkaľ, Ján Mezey

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Summary

Biofortification is a promising and practical strategy to raise the concentration of essential micronutrients in the edible parts of horticultural crops or their products. The small-plot experiment was part of a scientific study aimed at increasing of organic selenium in carrot juice. The small-scale experiment was part of a scientific study aimed at increasing of organic selenium in carrot juice. It was determined how fortification will affect the yields of selected carrot varieties, as well as the juice yield potential and, finally, the content of selenium in the juice. Foliar application of sodium selenate in a total amount of 50 mg/l (90 ml/m²) was carried out on five carrot varieties ('Kamaran F1', 'Naval F1', 'Vita Longa', 'Niagara' and 'Baltimore F1') in two dates of application. Each variety was monitored in the control (without selenium application) and selenized variant, while each variant had 3 repetitions (1 repetition = 3 m row). The laboratory part of the experiment was carried out in the Beverage Laboratory in the AgroBioTech Research Centre, SUA, Nitra. A one-time manual harvest of carrot roots was carried out on August 19th, 2021. After weighing and cleaning the roots, an average sample (1000 g) was prepared from each variety, variant and repetition, which was cut into smaller pieces and then juiced (by Magimix Le Duo Plus XL juicer, France). The carrot yields, juice yield potential in % was subsequently calculated from the measured data. Average yield values of carrot roots ranged from 60 t/ha to 108 t/ha in the control and from 64 t/ha to 110 t/ha in the selenized variant. When monitoring the total yield, selenization led to an increase in the value of the varieties 'Naval' by 27%, 'Baltimore F1' by 12% and 'Kamaran F1' by 2.26%. The juice yield potential ranged from 46% to 48%, since in the selenized variant were higher in case of varieties 'Baltimore F1' about 2%, 'Kamaran F1' about 4.5% and 'Naval' about 1%. A significant effect ($P \leq 0.05$) of foliar selenization on selenium content was recorded in all tested varieties. The highest value (0.1382 mg/kg) in the selenized variant compared to the control sample was achieved in the variety 'Kamaran F1'. Since the content of selenium in fresh juice after the application of foliar biofortification with selenate significantly increased without a negative effect on other tested parameters, all selected carrot varieties can be recommend as suitable for foliar selenization with sodium selenate in a total dose of 50 ml/l. Selenium is an essential element for metabolism and human health, and according to our results, foliar selenization of carrots represents a suitable form of selenium biofortification even when processing carrots into juice. It would take 3.5 – 4.9 dcl of selenized carrot juice ('Kamaran F1' variety) to get the Recommended Dietary Allowance (RDA) for women (50 µg) and for men (70 µg), as opposed to 5.66 - 7.92 l of non-selenized carrot juice.

Keywords: carrot, foliar selenization, fortification, selenium

Acknowledgements: This research was funded by grant: KEGA 018SPU-4/2020 and KEGA 004SPU-4/2022.

Influence of selected biostimulants on quantitative and qualitative parameters of pepper (*Capsicum annuum* L.)

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Summary

The aim of the research was to determine the influence of the application of selected commercial biostimulants on the cultivation of pepper (*Capsicum annuum* L.), cv. 'Kurtovszka kápia', in the conventional cultivation system in combination with nitrogen fertilization. An experiment with two variants and a control was set up according to the block design in three replications in a tunnel in the Botanical Garden of the Slovak University of Agriculture in Nitra. In the first variant, the preparation 'Energen Fulhum Plus' (0.5 L ha⁻¹) was applied 1 time before planting the seedlings by soaking the root ball and the preparation 'Energen Fructus Plus' (0.5 L ha⁻¹) was applied 2 times during vegetation by spraying the plants. In the second variant, the preparation 'Humix® Univerzál' (5 L ha⁻¹) was applied 3 times during cultivation (before planting by soaking the root ball and 2 applications during the vegetation period by spraying the plants). The harvest of fresh pepper fruits at full botanical maturity was carried five times, from August 2 to October 20, 2022. The results showed that the tested preparations had no positive effect on fruit weight and total yield of pepper fruits, as well as on the content of analysed nutritional parameters (total carotenoids and vitamin C). However, the application of the preparation based on humic acids ('Humix® Universal') significantly increased the content of refractometric and gravimetric dry matter of the pepper fruits.

Keywords: yield, carotenoids, vitamin C, refractometric and gravimetric dry matter

Acknowledgements: This research was funded by grant: KEGA 018SPU-4/2020 and KEGA 004SPU-4/2022.

Lamb's lettuce morphology and yield under different glass coverings

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Summary

The aim of this research was to determine the effect of nanoparticle-coated glass (Germanium (Ge), Titanium nitride, Ge + Silicon) as greenhouse cover on the morphology and yield of lamb's lettuce. The tested glasses differ in terms of total light transmittance and transmittance of wavelengths 450 and 650 nm. The transmittance of the nanoparticle-coated glasses ranged from 44.4 to 79.1% of the light transmittance of standard horticultural glass. Transmittance for 450 and 650 nm wavelengths ranged from 3.1 to 63.2% and from 39.9 to 77.1%, respectively. The air temperature and humidity sensor and a PAR sensor connected to the data logger were installed in the greenhouses. During harvest, conducted 30 days after planting, total biomass, number of leaves, mass of rosettes and mass of roots were determined. The highest values of all parameters were determined for lamb's lettuce grown under standard horticultural glass (plant biomass 2.65 g, rosette mass 2.53 g, root mass 0.12 g, 11.5 leaves, yield 1.265 kg m⁻²). In greenhouses covered with tested nanoparticle-coated glass, the highest values of monitored traits were obtained under glass coated with Germanium. Although a significantly lower yield was achieved under the tested nanoparticle-coated glasses, the use of glass with suitable structure has an advantage, especially in the warmer part of the year due to reduced light transmittance, as there is a lower temperature gradient compared to standard glass.

Keywords: *Valerianella locusta*, glass structure, nanoparticles, leaves number, rosette mass

Wild geophytes in Croatian vascular flora and its potential use in sustainable horticulture

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Summary

Geophytes, herbaceous plants with underground storage organs (bulbs, rhizomes, etc.), create an important part of Croatia's floristic wealth, about 14% of the total 5153 vascular plants. Over the last decades, interest in the use of wild plants in horticulture has increased. In this study, wild geophytes in Croatia were analyzed and selected those which are possible to be used as ornamental plants. The analysis revealed a total of 55 species and subspecies of wild geophytes (belonging to 34 genera and 19 families) with potential ornamental value. Of the 55 identified taxa (53 species and two subspecies) of wild geophytes, 48 species belong to the angiosperms, and seven to the ferns. Furthermore, it has been found that 33 taxa had rhizomes and 22 had bulbs. When the wild geophytes were analyzed in terms of flower color, the most common flower color is white (32%), followed by other colors (purple, pink, yellow, etc.). In conclusion, factors such as urbanization, tourism, and human impacts threaten the habitats of wild geophytes and it is very important to reduce these effects to a minimum level and at the same time ensure that the taxa are kept in place and out of place. In this context, the use of geophytes belonging to the wild taxa of Croatian flora will provide great contributions to sustainable horticulture, reduction of environmental problems and conservation of biodiversity.

Keywords: wild geophytes, ornamental plants, flower color, vascular flora, Croatia

Accumulation of stinging nettle polyphenols as a response to drought stress

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Summary

The hydroponic ebb and flow system is a modern technique most commonly used for growing leafy vegetables. It involves periodic flooding and draining of the nutrient solution, which allows controlled drought stress. Abiotic stress can positively affect the accumulation of plant polyphenols with high antioxidant activity. Stinging nettle is a perennial wild plant that can be consumed as a green leafy vegetable due to its nutritional value. The aim of this research was to determine the content of total phenolics (TP), flavonoids (TF), non-flavonoids (TNF) and antioxidant capacity of stinging nettle leaves grown in ebb and flow system under different irrigation intervals. The experiment was conducted in a greenhouse during the spring growing season. Plants were treated with a nutrient solution for one hour and then exposed to three different drought intervals of 24, 48, and 96 hours. During the irrigation interval of 48 hours, plants accumulated the highest amounts of TP ($400.21 \text{ mg GAE } 100 \text{ g}^{-1}$), about 23% more than when plants received no nutrient solution for 24 hours, and even about 30% more than when the plants were not irrigated for 96 hours. Also, the values of antioxidant capacity were highest during the 48-hour irrigation interval. The obtained results suggest that stinging nettle accumulates the most phenolic compounds during the 48 hours stress caused by the absence of nutrient solution

Keywords: abiotic stress, drought stress, ebb and flow hydroponic system, phenolic compounds, antioxidant capacity

Drought impact on kale landraces through agromorphological parameters

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Summary

Maintaining sufficient yield and quality is necessary to ensure food security under limited water availability. Drought can negatively affect the growth rate of *Brassica* species and consequently reduce yield. Kale (*Brassica oleracea* var. *acephala*) is traditionally cultivated on the Croatian coast. It is a leafy vegetable valued by scientists and consumers due to the presence of specialized metabolites such as glucosinolates (GLS), but also for its ability to withstand harsh cultivating conditions. The goal of this research was to test 6 kale landraces in their ability to withstand drought stress by evaluating their yield components. The kale landraces were cultivated in well-watered conditions, under moderate drought, and drought stressed conditions and planted at 3 different dates. Significant differences in yield were found among the landraces as well as different planting dates and drought intensity levels. Total GLS content differed by planting date and landrace, and a significant interaction between planting date and landrace was found for sinigrin. Kale landrace 'IPT415' had the highest yield among the tested landraces. Higher yield was achieved on the second planting date, while yields were not significantly different under well-watered and moderate drought conditions. Higher kale yield with less water use could be achieved through combining planting dates with moderate drought. Higher GLS content could be achieved through a selection of landraces and appropriate planting dates.

Keywords: *Brassica oleracea* var. *acephala*, abiotic stress, yield, GLS, planting dates

Traditional and modern yards' horticultural flora of Ludbreg Podravina

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Summary

Changes in the way of life and economy drastically change the use, arrangement and selection of plant taxa in country gardens. Traditional gardens are disappearing and with them valuable taxa of ornamental and useful plants, as well as local names and knowledge about them. The aim of this work is to study the size of the homeyard of the Ludbreg region, and to find out which plant taxa are grown on traditional and modern homeyards. The average size of the studied traditional homesteads is 1820 m², and their average built-up rate is 16.22%. They are smaller than the modern ones, whose average size is 2,646 m², and whose average built-up area is 16.80%. On the traditional home gardens, a total of 204 taxa from 72 families were recorded, with the most represented families being Asteraceae and Rosaceae, with 16 taxa each. There are 154 decorative taxa, 33 useful taxa, while 17 taxa are of mixed decorative-utilitarian purpose. A total of 293 taxa from 89 families were identified in the inventory of modern homeyards. The most numerous are the Asteraceae family with 23 taxa and the Rosaceae family with 22 taxa. There are 222 decorative taxa, 41 utilitarian taxa, and 30 decorative-utilitarian taxa.

Keywords: garden surface, ornamental plants, tradition, Varaždin County, vegetation

The influence of irrigation on growth and development of potted *Catharanthus roseus*

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Summary

The aim of this study was to analyze the morphological and anatomical responses of potted Madagascar periwinkle to deficit irrigation and to evaluate the possibility of producing commercial quality plants with less water use. Seedlings of Madagascar periwinkle (*Catharanthus roseus* (L.) G. Don 'SunStorm Red') were subjected to three irrigation treatments: container capacity (control), 80% of the control (mild deficit irrigation) and 60% of the control (moderate deficit irrigation). Deficit irrigation (DI) treatments caused a decrease in stem length and diameter, plant width, number of lateral branches with visible flower buds and the size of the leaf blade, but did not affect leaf thickness and the number of nodes formed before flowering. Leaf chlorophyll content index (CCI) increased with increasing water deficit and, in moderate DI treatment, the leaves were noticeably darker green than in the control plants. DI treatments also led to a decrease in the size of the stomata, on both sides of the leaf, and to an increase in the stomatal density on the abaxial side of the leaf. Water deficit did not considerably affect the start of flowering, but it led to a decrease in flower diameter and, in the moderate DI treatment, lowered the number of flower buds per plant. Mild DI did not significantly impair ornamental quality of the plants and therefore could be applied in production of Madagascar periwinkle to reduce water consumption.

Keywords: Madagascar periwinkle, deficit irrigation, ornamental quality, plant production

Agronomic potential and nutritional characterization of open-field cultivated purslane

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Summary

Purslane (*Portulaca oleracea* L.) is an annual herbaceous plant originating from India but is now common throughout the world. Due to its high nutritional value and ability to withstand drought and high salinity, it is becoming an interesting new agricultural crop. Its cultivation would contribute to the preservation of biodiversity and the environment and would fit perfectly into the framework of sustainable agriculture, since it is a wild species considered by farmers as a weed. The aim of this research was to determine the differences in agronomic and nutritional behavior of two purslane accessions based on yield parameters (yield, fresh and dry weight of leaves and stems, dry matter) and antioxidant capacity under field growing conditions. Yield ranged from 1800 to 3400 g m⁻² at a planting density of 10 plants m⁻². No statistically significant difference was found between the two accessions in the observed agro-morphological parameters except for stem dry weight. In the applied antioxidant capacity analyses, the only significant difference between the accessions was detected by the ORAC method, with the 'IPT478' accession having a higher antioxidant capacity. Due to its high growth rate and ability to withstand several abiotic stressors, purslane is a potential candidate for field cultivation with yields up to 34 t ha⁻¹, according to this study. The antioxidant capacity results also show purslane's potential as a highly nutritious crop.

Keywords: *Portulaca oleracea* L., yield, antioxidant capacity, dry matter, vegetable

Morphological properties and yield of chickpea depending on rhizobial inoculation and intercropping with black cumin

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Summary

In order to evaluate the beneficial effects of intercropping chickpea with black cumin and inoculation with indigenous rhizobia, an experiment was conducted in 2022 at the Faculty of Agriculture in Zagreb. The two-factorial research was set up according to the randomized block design with three replications. The 1st factor was intercropping: without (WOI) and with (I); the 2nd factor was inoculation: without (WOS) and with the Spanish reference strain (ISC11) and the Croatian indigenous strain (47b). Chickpea height, number of fertile branches and pods per plant, and grain yield were determined. Intercropping had no effect on the observed traits and yield. Inoculation with strain 47b resulted in a significantly higher number of fertile branches and pods (10.2 and 40.8) than the reference strain (8.3 and 34.9). The smallest plant height (43.7 cm) achieved by the I×ISC11 interaction was significantly smaller than by I×47b and I×WOS (47.4 cm), WOI×ISC11 (47.2 cm), and WOI×47b (46.7 cm). Interaction I×47b had the highest number of fertile branches (10.7), statistically equal to the values of 10.2, 9.6, and 9.5 obtained by I×WOS, WOI×47b, and WOI×WOS, respectively. The highest grain yield (0.329 kg m⁻¹) obtained by WOI×47b was significantly higher than that of WOI×WOS (0.245 kg m⁻¹) and statistically equal to the yield of the other interactions. Based on the above, further research should be focused on testing the effective strain 47b and some perspective indigenous strains but without intercropping with black cumin.

Keywords: *Cicer arietinum*, *Nigella sativa*, indigenous rhizobia, fertile branches, pods

A green strategy to isolate bioactive extracts from wild edible plants

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Summary

The main objective of this research was the extraction of bioactive compounds from different parts of two wild edible plants, *Diploaxis eruroides* (pods, leaves, stems) and *Oxalis pes-caprae* (flowers, leaves, stems), by green strategies and encapsulation of the extracts. The flowers of *Oxalis pes-caprae* showed the highest content (about 750 mg GAE g⁻¹) of total phenolics. The pods of *Diploaxis eruroides* showed the highest total phenolic content (47 mg GAE g⁻¹) compared to the leaves and stem. All encapsulated samples showed promising results regarding the release of bioactive compounds. Another important factor of the encapsulations for their practical application is the swelling capacity, and the pods of *Diploaxis eruroides* samples showed the highest percentage of swelling (217%). However, for *Oxalis pes-caprae*, the results were very similar for flowers and stems (124% and 127%, respectively). In conclusion, these encapsulated plant extracts showed potential properties so they could be used as promising candidates for application in various fields such as food, cosmetics or agriculture.

Keywords: encapsulation, total phenolics content, functional food, high-added value ingredients

Sustainable bioconversion of wetland plant waste into wealth: proximate, biochemical and nutrient characterization of cultivated *Pleurotus florida* mushroom

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Summary

Abundant biomass growth of aquatic macrophytes in wetlands is one of the top concerns affecting their residing biota. In addition, wetland biomass degenerates within the wetlands which cause remixing of nutrients and emits greenhouse gases. Therefore, it is important to find sustainable methods to utilize aquatic macrophyte biomass to avoid several environmental problems. Considering this problem, the present study deals with the utilization of biomass of three aquatic macrophytes, including lake sedge (CL: *Carex lacustris* Willd.), water hyacinth (EC: *Eichhornia crassipes* Mart. Solms.) and sacred lotus (NL: *Nelumbo nucifera* Gaertn.) to produce oyster mushrooms (*Pleurotus ostreatus* var. *florida*). For this purpose, different combinations of wheat straw (WS: as control) and macrophyte biomass such as control (100% WH), CL50 (50% WH + 50% CL), CL100 (100% CL), EC50 (50% WH + 50% EC), EC100 (100% EC) and NL50 (50% WH + 50% NL), NL100 (100% NL) were used for *P. florida* cultivation under controlled laboratory conditions. The results showed that all selected waste combinations promoted spawning and growth of *P. florida*. In particular, the highest significant ($p < 0.05$) growth, yield, bioefficiency, proximate, and biochemical parameters were reported using WH substrate followed by CL50, NL50, and EC50 treatments, which correspond to the reduction efficiency of substrate parameters, respectively. Therefore, the findings of this study reveal that the biomass of selected aquatic macrophytes can be effectively used for sustainable mushroom cultivation while minimizing the risk of self-degeneration.

Keywords: waste management, sustainable agriculture, mushroom cultivation, super foods, pollution reduction

Yield and antioxidant activity of turnip after application of soil biostimulant 'Agriful'

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Summary

Turnip is one of the less known root vegetables in the Slovak Republic. The aim of this study was to evaluate the effect of soil biostimulant 'Agriful' on yield parameters and antioxidant activity (AOA) of turnip roots. The field experiment with four turnip cultivars ('Amelie F1', 'Oasis F1', 'Purple Top White Globe', 'Tonda a Colletto Viola') was laid out in 2021 at the Slovak University of Agriculture in Nitra. The soil biostimulant 'Agriful' (based on humic and fulvic acids) was applied four times in two-week intervals during the turnip vegetation period. Roots were harvested on 4th October and successively classified into quality classes according to UNECE standard FFV-59. The AOA of turnip roots was determined in lyophilized samples using 3 different methods (DPPH, FRAP and ABTS). Statistical analysis revealed significant differences in yield and average root weight between the control and 'Agriful' treatment in all tested turnip cultivars. The increase of turnip yield after 'Agriful' application was ranged from 22.6 to 51.3% ('Amelie F1'). The application of 'Agriful' resulted in a decrease in the percentage of non-standard quality roots and an increase in the percentage of 1st quality. After the 'Agriful' application, the AOA of turnip was higher compared to the control variant, with the exception of DPPH AOA in 'Oasis F1' (decrease of about -5.4%). According to experimental results, it is evident that cultivar, as well as biostimulant application, are significant factors influencing on the yield and AOA of turnip.

Keywords: *Brassica rapa* L. ssp. *rapa*, cultivar, biostimulant, 'Agriful', roots

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Session **5.2** Book of Abstracts
Viticulture, Enology and Pomology

58 hrvatski

18 međunarodni
simpozij
agronoma

Zbornik sažetaka

Vinogradarstvo, vinarstvo i voćarstvo

The genetic and geographical groups of grapevine varieties discriminated by polyphenolic and volatile profiles

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Summary

Grapevine germplasm is highly variable and classified into geographical groups. These classifications were recently confirmed by genetic studies, and further classified into genetic-geographic (GEN-GEO) groups. Secondary metabolite, namely polyphenolic and volatile organic compounds (VOCs), have crucial role in winemaking industry due to their influence on quality, color, and sensory properties of wine. The aim of the research was to investigate polyphenolic and volatile profiles of 50 grapevine varieties from different GEN-GEO groups: C2 (Italy, France), C7 (Croatia), C8 (Spain, Portugal). The most abundant classes of polyphenols were anthocyanins, flavan-3-ols and flavonols, while carbonyls alcohols and sesquiterpenes were the most abundant classes of VOCs. Using discriminant analysis, the GEN-GEO groups were clearly separated by their polyphenolic and volatile profiles. In the case of polyphenolic profiles, compounds contributing the most to the discrimination of groups belong to classes of hydroxycinnamic acids, flavan-3-ols, and flavonols. Regarding the discrimination based on volatile profiles, GEO groups were discriminated by their overall volatile profile. C2 group contains higher amounts of carbonyl compounds and alcohols, C8 group contains higher amounts of sesquiterpenes and acids. Group C7 is characterized by low content of VOCs. This data demonstrates that geographical origin, combined with genotype, also influences the overall polyphenolic and volatile profiles of grapevine varieties.

Keywords: secondary metabolites, grapevine varieties, GEN-GEO groups, discriminant analysis

The influence of heat stress on the pomological, physiological and chemical characteristics of 34 apple varieties

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Summary

The joint project “APPLERESIST” of the Faculty of Agrobiotechnical Sciences Osijek and the Croatian Agency for Agriculture and Food, Center for Fruit and Vegetable Growing, planted an experimental orchard with 34 varieties of apples. This project will determine the genetic resistance of an assortment of apples to heat and drought stress, as well as the possibilities of adaptation to climate changes, with the recommendation of a certain assortment for cultivation in the continental part of the Republic of Croatia. The project is carried out at two locations: the FAZOS test site in Tenja (Osijek) and the HAPIH test site in Donja Zelina. In the fall of 2021, two-year-old apple seedlings were planted with the following varieties: Collina, Allegro, Galaval, Gala Schniga Schnico, Gala Galaxy, Gala Schnitzer Schniga, Gala Dark Ann, Santana, Freya, Rozela, Orion, Wiltons Star Red Jonaprince Select, Bay 3341, Merkur, Golden Delicious Smoothee, Crimson Crisp Coop, Jonagold Novajo, Sirius, Golden Delicious Reinders, Reglindis, Opal, Jeromine, Red Idared, Super Chief Sandidge, Gold Rush Coop, Red Topaz, Topaz, Pinova, Karneval, Roats King Red Delicious, Braeburn Lochbuie, Braeburn Aporo Mariri Red, Granny Smith and Fuji Kiku. Drip irrigation systems and anti-hail nets were installed in the plantations. The distance between the rows is 3.2 m, and within the row is 0.8 m. The fruits for determining the pomological, physiological and chemical characteristics were harvested successively as the varieties ripened from July 21 to October 19. Thirty average fruits of each variety were analyzed at both locations, and even though it was an extremely dry year with temperature extremes, preliminary results were obtained as follows: Donja Zelina experiment; The average weight of the fruits was from 113.00 g (Gala Schnitzer Schniga) to 260.55 g (Freya), fruit height was from 51.74 mm (Galaval) and 74.87 mm (Granny Smith), the circumference ranged from 63.10 mm (Gala Schnitzer Schniga) to 87.10 mm (Freya). Fruit hardness ranged from 5.01 kg/cm² (Collina) to 9.82 kg/cm² (Gold Rush Coop). Total soluble dry matter was determined using refractometer and was between 12.33 Brix⁰ (Collina) – 17.75 Brix⁰ (Fuji Kiku). Total titration acids, expressed as malic, ranged from 12.92 g/l (Super Chief Sandidge) to 30.55 g/l (Bay 3341). The average pH was from 3.35 (Bay 3341) to 4.62 (Super Chief Sandidge). Trial site Tenja; Average fruit weight was from 137.38 g (Allegro) to 297.75 g (Santana), the height was from 55.77 mm (Allegro) to 80.74 mm (Santana), and the circumference from 70.50 mm (Allegro) to 92.50 mm (Sirius). Fruit hardness was from 5.10 kg/cm² (Santana) to 9.94 kg/cm² (Crimson Crisp Coop). Total soluble dry matter was from 12.41 Brix⁰ (Allegro) – 17.43 Brix⁰ (Fuji Kiku), and total titration acids, expressed as malic, were from 12.92 g/l (Super Chief Sandidge) to 30.55 g/l (Bay 3341). The average pH of the fruits ranged from 3.45 (Red Topaz) to 4.61 (Super Chief Sandidge).

Keywords: apple, varieties, heat stress, pomology

Influence of rhizobacterium on the vegetative activity of one-year-old MaxMa 14 rootstocks

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Summary

The influence of the rhizobacterium *Azospirillum brasilense* on the vegetative activity of MaxMa 14 (*Prunus mahaleb* x *Prunus avium*) rootstocks, a widely used sweet cherry rootstock in semi-intensive orchards, was determined. The field experiment was conducted on one-year-old micropropagated rootstocks in 3L pots, arranged to randomized block design in 3 repetitions, and 15 pots in row per each repetition. Vegetative growth promoter AZOS® that contains single rhizobacterium *A. brasilense* was added by watering to plants in concentration of 0 g/l (T0 control), 0.8g/l once a month (T1) and 0.8g/l every two months (T2) in controled time intervals. Plant height, trunk cross-section area (TCSA), shoot number and chlorophyll content index (CCI) were observed as factors of vegetative activity. Also, vegetative growth of 3 individual shoots and number of their internodes was measured three times during vegetation. Based on the obtained research results, it was determined that the use of *A. brasilense* by watering rootstock seedlings affected vegetative activity. Plant height, TCSA, CCI, shoot length and internode number were influenced by *A. brasilense* while shoot number wasn't influenced by rhizobacteria. In conclusion, watering MaxMa 14 rootstocks with *A. brasilense* could be useful for obtaining quality grafting material.

Keywords: rhizobacterium, rootstock, sweet cherry, vegetative activity, watering

Influence of biofortification with selenium and zinc on qualitative properties of must and wine cv. Cabernet Sauvignon

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Summary

The research was conducted during 2021 at the Faculty experimental field Mandićevac, part of vinegrowing area Đakovo. The aim of the research was to determine the influence of biofortification with selenium and zinc on some qualitative properties of must and wine cv. Cabernet Sauvignon. Experiment was set up with 2 treatments in 6 replicates by completely randomised design. Biofortification with selenium and zinc was carried out in the flowering and veraison stage. After the preparation of must and wine samples, *in vitro* digestion simulation was carried out according to Minekus et al. (2014). Total and bioavailable concentrations of zinc and selenium in must and wine samples were determined using ICP-OS and ICP-MS techniques. In addition, standard physico-chemical analyses were performed on must and wine samples. Statistical data processing was performed using the SAS Enterprise Guide 7.1 program, and the t-test was used to determine differences between the tested treatments. The conducted t-test determined the significant influence of biofortification with zinc and selenium on the total concentration of zinc in wine, the bioavailable concentration of zinc in must and the bioavailable concentration of selenium in wine. The influence of biofortification with zinc and selenium on the investigated qualitative properties has not been determined.

Keywords: biofortification, selenium, zinc, must, wine

Influence of harvest date on Malvazija istarska volatile composition as revealed by HS-SPME-GC×GC-TOF MS

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Summary

Anecdotal observations have shown that there is often incoherence between sugar accumulation and evolution of grape flavour during later ripening stages. For this reason, an experiment with Malvazija istarska (*Vitis vinifera* L.) was conducted to investigate the relationship between harvest date and changes in the abundance of wine volatile aroma compounds. Malvazija grapes were harvested and subsequently vinified at three harvest dates (H1 – H3) separated from each other by approximately 10 days, with grape quality parameters at each date suitable for production of standard still white wines. Wine volatile aroma compounds were analysed by comprehensive two-dimensional gas chromatography with time-of-flight mass spectrometry combined with conventional one-dimensional GC/MS and were statistically elaborated. Various terpenes and alcohols were found in the highest concentration in wines from the grapes harvested at H3. Most of fermentation aroma compounds showed no significant differences. The exceptions were ethyl-2-methylbutyrate, ethyl propanoate and isobutyl decanoate found in the highest concentrations in wines from H3 and isopropyl acetate and isoamyl decanoate in wines from H1. The obtained results confirmed that by choosing the harvest date it is possible to significantly influence the composition of volatile aroma compounds in white wine.

This study was funded by Croatian Science Foundation under the projects IP-2020-02-4551 and DOK-2021-02-5500.

Keywords: harvest date, volatile aroma compounds, Malvazija istarska, two-dimensional gas chromatography, one-dimensional GC/MS

Effect of clarification with various types of bentonite on proteins, phenols, and aromas in Malvazija istarska white wine

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Summary

The aim of this study was to investigate the effect of clarification with various types of bentonite during and after fermentation on proteins, phenols, and volatile aromas in Malvazija istarska white wine. Proteins were analysed by 2D proteomics with ion exchange chromatography and SDS-PAGE separation followed by MALDI-TOF/TOF identification, while phenols and volatile aromas were determined by HPLC-DAD and HS-SPME-GC/MS, respectively.

In the first experiment, four commercial bentonites were dosed late in fermentation for partial protein stabilization, while control received no bentonite. Varying efficacy regarding protein stability was observed, depending on bentonite type. Various bentonites exhibited differential and preferential affinity towards particular proteins, as well as variable, mostly positive side-effects on the contents of particular odoriferous esters and antioxidant phenols.

In the second experiment, wines were clarified to complete protein stability prior to bottling by nine commercial bentonites. Significant differences in the required doses were determined. Bentonites showed diverse effects on wine chemical composition, but each deprived the wine of variable proportions of most of the volatile esters.

The obtained results clearly pointed to the benefits of the application of bentonite for partial wine stabilization during fermentation.

This study was funded by Croatian Science Foundation under the projects UIP-2014-09-1194 and IP-2020-02-4551.

Keywords: clarification, bentonite, pathogenesis-related proteins, fermentation, Malvazija istarska wine

Optimization of preculture medium for *in vitro* microcuttings in the cryopreservation procedure of the grapevine cultivar 'Graševina'

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Summary

Grapevine (*Vitis vinifera* L.) is one of the oldest agricultural species. Wide application of this culture in the economy makes it one of the most important agricultural fruit culture in the world. Republic of Croatia is important gene center for native, as well as for introduced cultivars of grapevine, so the viticulture's aim is to conserve and revitalize its cultivation. Cryopreservation is the most efficient procedure for the conservation of the plant material. In the cryopreservation procedure different preculture of microcuttings, cryoprotectants and steps during freezing can be toxic and make the stress within cultivars of grapevine. The aim of this study is the optimization of preculture medium for preculture of microcuttings with addition of antioxidants (salicylic acid) with the purpose of the successful growth of shoot tips of the cultivar 'Graševina'. The study was made on microcuttings of cultivar 'Graševina', planted on half-strength MS medium, with or without cytokinins, benzylaminopurine and different concentrations of salicylic acids (0, 0.1, 0.5 and 1 mMol). The highest percentage of shoot microcuttings was achieved on the medium without the salicylic acid and BAP (68, 38%), and the lowest on the medium with addition of salicylic acid in concentration 0.1 mMol and supplement of 1μmol BAP (35, 00%). Microcuttings cryopreservation of cultivar 'Graševina' reached the highest results of regeneration in controlled explants (15%) in comparison on the freezing ones (10%). Given results are implying that some additional studies should be done for successful cryopreservation of this cultivar.

Keywords: *Vitis vinifera* L., cultivar 'Graševina', preculture of microcuttings, antioxidants, salicylic acid, regrowth

Olive leaf mineral and total phenolic concentrations: The effect of different cultivars

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Summary

The olive leaf is a valuable source of different phytochemical compounds which play crucial role in plant defence against different pathogens or can be used for range of different nutraceuticals. Nevertheless, the olive cultivars can be differed by their leaf mineral or total phenolic content but possible link between them is still unequivocally established.

Thus, the aim of this work was to investigate the difference in minerals and total phenolic concentrations in olive leaves of four Croatian autochthonous cultivars (Drobnica, Levantinka, Lastovka, and Oblica) and Italian Leccino as one of the most common allochthonous olive cultivar in Croatian olive orchards. The trial was selected as a completely random design (n = 3) during March pruning period in 2018.

Selected cultivars had shown significant differences in leaf potassium (K), magnesium (Mg), iron (Fe), zinc (Zn), manganese (Mn), and boron (B) concentrations. Total phenolic concentration was significantly different between Levantinka cv. (8022 mg/100 g DW), Oblica cv. (7296 mg/100 g DW), Leccino cv. (6570 mg/100 g DW), Lastovka cv. (5844 mg/100 g DW) and Drobnica cv. (4233 mg/100 g DW) respectively. A strong positive correlation was noticed for total phenolic and Mg leaf concentrations ($r=0,72$, $p<0.01$).

Due to mineral and phenolic differences selected Croatian olive cultivars should be tested in future Mg plant nutrition trials.

Keywords: Levantinka, Oblica, Leccino, Lastovka, Drobnica.

Bunch load as a factor of the yield and quality of Victoria and Ribier table grape varieties

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Summary

The research was conducted over the period of three consecutive years (2014-2016) on the production plantations of Victoria and Ribier table grape varieties in Gevgelija-Valandovo vine-growing region. After the blooming phenophase, the number of bunches (crop load) was reduced. Three bunch thinning variants were applied: B1 (thinning to 4 bunches/m²), B2 (thinning to 5 bunches/m²), and B3 (thinning to 6 bunches/m²). The variants were compared with standard (St) where no thinning of bunches was applied. During the vegetation, standard agrotechnical and ampelotechnical measures were implemented for all variants, including the standard.

The average bunch and berry mass in both varieties increase with increasing bunch reductions. In the Victoria variety, the largest individual bunch mass of 689,7 g and berry mass of 8,8 g was obtained in the B1 variant, while the lowest bunch mass of 603,3 g and berry mass of 8,0 g was obtained in the standard variant. In the Ribier variety, the highest bunch mass was obtained in variants B1 (445,7 g) and B2 (432,7 g), a mass that is statistically significantly different in relation to the average mass in the variants St and B3. The largest amount of total yield in the Victoria variety was obtained in the variants St (2,93 kg/m²) and B3 (2,91 kg/m²). The B3 variant is also characterized by a larger amount of packed grapes with 2,54 kg/m². The total yield in the Ribier variety shows approximately equal results in the three of the variants i.e. St (2,76 kg/m²), B2 (2,74 kg/m²), and B3 (2,79 kg/m²), but the amount of total packed grapes is highest in B2 variant (2,31 kg/m²), a quantity that is statistically significantly different compared to the other variants. In terms of organoleptic evaluation, the best grades and category of excellent quality were obtained for variants B1 and B2 in both of the tested varieties.

Keywords: table grapes, bunch thinning, bunch and berry mass, yield, quantity of packed grapes.

Results of marker-assisted selection for downy and powdery mildew resistance in grapevine breeding program

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Summary

Modern viticulture in Croatia and the world is based mainly on varieties sensitive to various diseases and pests, resulting in the unsustainably large amounts of pesticides used in grape production. The sustainable development of viticulture in the future will only be possible by increasing the resistance of the grapevine through the development of new varieties resistant to diseases and abiotic stresses caused by climate change. Breeding programs have been launched in the leading wine-growing countries to develop new varieties with resistance to diseases (resistant varieties) using classical breeding and selection supported by genetic markers (MAS - Marker Assisted selection). In 2015, a breeding program was started in Croatia, whose long-term goal is the development of resistant varieties of grapevines with stable and durable resistance, using autochthonous germplasm in combination with different sources of resistance that are present in the resistant varieties developed so far suitable for growing in Croatia. This research presents the results of the application of the molecular markers with the aim of the detection of resistant gen-loci in the offspring obtained from two crossings: 1st native varieties 'Grk' and 'Dišeća ranina' with the resistant variety 'Panonia' and 2nd crossing the selected genotypes from first crossing with genotype SK 00-1/8. Resistant gen-loci which were analyzed in progeny are RpV3, RpV12, and Ren3 (from 'Panonia'), and Run1 and RpV1 (from SK 00-1/8). The obtained results showed that it was possible to detect several genotypes with pyramided resistance, i.e., multiple gen-loci related to resistance to grapevine mildews.

Keywords: grapevine, breeding, marker-assisted selection, resistant varieties

Polyphenol content and antioxidant activity in blackberry (*Rubus fruticosus* L.) of various origin

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Summary

The research aimed to determine the content of bioactive substances in blackberry (*Rubus fruticosus* L.) samples from different regions of Slovakia and the Netherlands and other origins, namely forest, domestic production, and commercial production. The research objective is divided into three sub-objectives: (i) determination of the total polyphenol content; (ii) determining the total anthocyanin content; (iii) determination of antioxidant activity. The highest contents of polyphenols in the samples were in fresh forest blackberries of Slovak origin (Starý Smokovec and Hincovce) with values of 3948.89 mg kg⁻¹ and 3752.52 mg kg⁻¹ while the highest content of anthocyanins was determined in fresh blackberry from the Veľká Lomnica (2804.07 mg kg⁻¹). In comparison, samples of fresh blackberries from Slovakia had a higher content of polyphenols and anthocyanins than samples from the Netherlands. The forest samples were characterized by the highest total content of polyphenols among all samples, while the highest anthocyanins content characterized the samples from domestic production. The forest sample from the location Starý Smokovec had the highest antioxidant activity (89.3% DPPH). In the assessment of antioxidant activity, there was no significant difference between the samples. Samples of blackberry from the wild and domestic production achieved similar results. This study proved that Slovakia is a country with excellent climatic and growing conditions for blackberry, and fresh blackberry from domestic and natural conditions shows significantly better values of biologically active substances. Determining the content of polyphenols and anthocyanins and antioxidant activity in blackberry are beneficial evidence for their inclusion in the diet as biologically active components of healthy human nutrition in the prevention of modern civilization diseases (i.e. cardiovascular diseases).

Keywords: Blackberry, polyphenols, anthocyanins, DPPH

Effect of grow tubes on vegetative growth of planted Marselan vines

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Summary

The use of plastic grow tubes on vegetative growth of the shoot – length, diameter, weight and dry matter have been presented in the paper. The investigations have been conducted in the vineyard of the Experimental Station Radmilovac of Faculty of Agriculture (E 44° 45' 15", N 20° 34' 56"). The trial has been established as two factorial with 30 vines in both variants, with grow tubes (Gt) and control (C). The grow tube was installed after planting of Marselan vines and standard agro and ampelotechnic operations have been performed during vegetation except additional removing of the weeds that were emerged inside grow tubes. The grow tubes had no effect on differences in shoot length (Gt=78,6 i K=69,4 cm); content of dry matter (Gt=56,3 i K=55,6 %) and shoot weight (Gt=53,19 i K=58,1 g), while differences between shoot diameter were significant (Gt=7,48 i C=9,25 mm). Obtained results indicate that using of grow tubes has no significant effect on the elements of vegetative growth of the planted vines. Considering the costs of purchasing and installing grow tubes and the need for additional engagement of workers to remove weeds around the protected vines, their application did not prove to be expedient during the 2021 growing season in the agro-ecological conditions of Radmilovac vineyards.

Keywords: grow tubes, shoot length, diameter, weight, dry metter.

Chemical composition of fruit wines from Croatia

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Summary

Fruit wines, in addition to enriching the offer of drinks on the market, are also a valuable dietary source of minerals, antioxidants and phytonutrients, especially polyphenolic compounds. Polyphenols represent an important group of compounds that influence not only the organoleptic characteristics of wine, color, taste, astringency and bitterness, but also physiological properties that have a positive effect on human health. Therefore, the aim of this study was to determine the chemical composition, chromatic parameters and antioxidant capacity of 20 commercially available fruit wines from Croatia produced from blackberry, chokeberry and cherry. The values of total polyphenols ranged from 819,00 mg/L GAE to 2442,38 mg/L GAE, which is equal or slightly lower compared to the content of total polyphenols in red wines. Wines produced from chokeberry (A2, A4, A5 and A3) and cherry (V4) stood out with the highest content of total polyphenols. The highest color intensity was measured in cherry fruit wines, V3 and V4. The values of antioxidant capacity ranged from 11,19 mmol/LTE to 42,25 mmol/LTE and were correlated with the values of total polyphenols. The antioxidant capacity of blackberry, chokeberry and cherry fruit wines is consistent with the antioxidant capacity of red wines, measured by the same method in previous research.

Keywords: fruit wines, polyphenolic compounds, chromatic parameters, antioxidant capacity

Nitrogen nutrition in olive orchards of Central Dalmatia

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Summary

The aim of the present study was to investigate the effect of fertilization practices, especially with nitrogen (N), on N leaf status, fruit and oil yield, and oil quality parameters in orchards of Central Dalmatia. Questionnaires done by more than 100 olive growers revealed that fertilization practices vary widely. Based on the data obtained, 22 orchards with most prevailing practices in N application were selected for detailed soil, leaf and fruit analyses. The samples were taken from trees of the Oblica cultivar. N concentration in leaves averaged 1.69% and ranged from 1.09 to 2.36% and correlated mainly with the intensity of agrotechnical practices and fertilizer use expressed in the questionnaires. Based on results from 22 orchards, an additional subselection of 9 orchards with low, optimal and high N leaf concentration was made. Hand-picked olives had on average 15.5% of oil, fruit N concentration ranged from 0.43 to 0.71%; with an average of 0.57%. The results showed correlation between N level in leaves and fruits, which was more pronounced in leaves with optimal N status. It was also found that N concentration in fruits was higher with lower K concentration in leaves. Significant negative correlation was found between N content in fruits and phenolic content in virgin olive oil, with increased N leading to sharp decrease in phenols in oil. As a basis for future studies on N nutrition in olive, fertilization experiment with three N supply levels was established on the cultivar Oblica.

Keywords: soil and leaf analyses, oil quality, phenolics, fertilization practices

Fruit quality of different sour cherry varieties (*Prunus cerasus* L.) in Eastern Slavonia

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Summary

During the growing season in 2022, the fruit quality of six cherry varieties was monitored in the cherry orchard in Jarmina; Oblacinska, Erdy botermo, Debrečen botermo, Sumadinka, Uifehert and Favorit. The orchard is five years old and the rootstock is *Prunus avium* L. The fruits are harvested at technological maturity. Pomophysical and chemical analyzes of fruit and seed weight, soluble dry matter and total acids were carried out in the laboratory. Data of yield and trunk cross-section area were also measured, from which the yield efficiency (YE) was calculated. Inflorescences and fruit set were counted and determined on the bearing branches with a total length of two meters, from which the fruit set and the index of the density of the inflorescences were calculated. The highest fruit set, yield and yield efficiency was recorded in the variety Sumadinka and the lowest in Favorit. The Oblacinska variety had the lowest fruit weight, and Erdy Botermo the highest. The lowest seed weight was recorded for the Uifehertska variety and the highest for Sumadinka. The variety Uifehert had the highest content of soluble dry matter, and Sumadinka the lowest. Total acids were the highest in the Oblacinska variety, and the lowest in Erdy Botermo. The quality of cherry fruits is mostly determined by the selection of the variety in production. Due to the very short harvest period of the Oblacinska sour cherry as the main variety in industrial processing, solutions are also being sought today regarding the introduction of an additional varieties in the production of industrial sour cherries.

Keywords: sour cherry, fruit, variety, quality

Effects of different irrigation regimes on vegetation indicators and qualitative characteristics of Teran grape (*Vitis vinifera* L.)

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Summary

Drought and the increase in air temperatures as consequences of climate change are directly reflected in the quantitative and qualitative characteristics of grapes. The objective of this study was to investigate in *Vitis vinifera* L. cv. Teran the responses of vegetation and grapes to different irrigation regimes: (V0) rainfed irrigation, (V1) irrigation up to 25% of estimated evapotranspiration (ET_c), (V2) irrigation 50% ET_c, (V3) irrigation 75% ET_c, (V4) irrigation 100% ET in 2022. The following indicators were monitored: vegetation indicators (NDVI) by remote sensing and sugar and acidity of grapes, which were measured at regular intervals to monitor ripening dynamics. Each experimental treatment consisted of three experimental replicates and each replicate included 20 vines. Must was obtained from the whole berry sample in which sugar content (Brix), total acidity (g/L expressed as tartaric acid) and pH were determined. Mean NDVI values ranged from 0.84 to 0.87, and the difference between V0 and all irrigated treatments was statistically significant. Of all the irrigated treatments, significantly lower NDVI values were measured in V1 and V2 than in V3 and V4, and grape analyzes showed significant differences between the irrigated and V0 treatments. Treatment V3 had higher pH and sugar content during the season than the other treatments. The lower acidity was found in treatment V3.

Keywords: precision irrigation, vineyards, remote sensing, water stress, grape

Pomological and physicochemical properties of traditional apple varieties from Koprivničko-Križevačka County

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Summary

Apple is known as one of the most widespread fruit species in the world. Despite a large number of varieties, only a few economically important varieties and their clones are represented in intensive production, while the traditional varieties are neglected. To highlight their potential, the possibility of growing native and traditional varieties of different fruit species has been recently intensively investigated. This work aimed to determine the pomological and physicochemical properties of the fruits of eight traditional apple varieties ('Ananas Reneta', 'Gospoinjača', 'Grofova', 'Imperica', 'Muškatna Mirisava', 'Boskop', 'Senabija' and 'Slastica') grown at the experimental site of the Križevci College of Agriculture. The results indicated that the investigated properties were significantly dependent on the traditional varieties. The largest fruits were found in the variety 'Grofova', and the smallest in the variety 'Muškatna mirisava'. The hardest fruit had the variety 'Ananas Reneta', and the softest 'Senabija', which had the highest total acid content. The highest content of total soluble solids content was found in the variety 'Boskop', and the lowest in the variety 'Imperica'. The fruits of the variety 'Slastica' had the lowest content of total acids and the highest pH value. The highest number of healthy seeds was found in the variety 'Muškatna mirisava', whereby 'Boskop' had the lowest number and seed mass. Traditional varieties are increasingly appreciated by consumers but occupy a small portion of the market. Therefore, it is important to conduct systematic research and encourage the cultivation of certain traditional apple varieties whose fruits are characterized by good quality and attractive taste.

Keywords: native varieties, apple, fruit quality, potential, cultivation

Which factor contributes most to the typicality of wine? A Slovenian case study

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Summary

By definition, vitivincultural terroir is a concept that refers to an area where a collective knowledge of the interactions between the identifiable physical and biological environment and applied vitivincultural practices develops, giving distinctive characteristics to the products coming from that area. Recently, there have been a number of approaches to underpin terroir with data and ways to distinguish between a conceptual and a perceptual approach. Thus, the objective of our case study was to deconstruct the influence of measurable parameters on grape yield and quality, wine aroma compounds, and sensory quality: namely, vineyard location, yield regulation by grape thinning, and type of alcoholic fermentation (inoculated vs. spontaneous). We have confirmed analytical markers that can distinguish thinning practice from control without yield regulation: wine TDM and pH, cis-3hexen-1-ol and 2-phenyl-ethyl-acetate. On the other hand, the location with its characteristics and the type of alcoholic fermentation significantly affected many more variables: standard wine parameters (wine TDM and pH, alcohol content), concentrations of aroma compounds (varietal thiols, higher alcohols and esters), and sensory quality measured by overall and attribute difference tests. We confirmed that both conceptually important factors (location with soil and climatic parameters) and perceived presentation (oenological practice) of typicality significantly influenced wine quality.

Keywords: grapevine, yield, yeasts, terroir, PDO wines



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agronoma

Zbornik sažetaka

Ratarstvo

Effect of period of application and amount of nitrogen fertilizer on the protein content of wheat grain

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Summary

Global changes caused by the coronavirus and the war in Ukraine have caused an increase in the prices of mineral fertilizers, and there is an increasing need to optimize the costs of agricultural production. The aim of the research was to determine which method of applying nitrogen fertilizers gives the best results in terms of yield and protein quality in winter wheat grains. In the growing season 2021/2022, field research was conducted on the yield and seed quality of winter wheat with different periods and amounts of Nitrogen fertilization application. The experiment was set up according to a RCBD scheme in CAAF Osijek (N 45°31', E 18°40') with seven variants of nitrogen fertilizer application in four repetitions with the applied standard agrotechnic. The research results showed that there was no statistically significant difference between the treatments on yield. The reasons can be found in the extreme drought during the spring part of the vegetation and the impossibility of adopting the applied nitrogen fertilizer. The protein content shows, a statistically significant difference between variants on the level $P < 0.05$, the treatments 5 (150 kg KAN/ha in tillering, 100 kg KAN/ha in stem elongation, and foliar 27 kg N/ha in heading) and 7 (150 kg KAN/ha in tillering and foliar 40.5 kg N/ha in heading) had higher values compared to treatment 1 (150 kg KAN/ha in tillering). The obtained research results will contribute to the correct selection of the time and amount of nitrogen fertilizer application on winter wheat at the Osijek location.

Keywords: wheat, yield, protein, starch, nitrogen fertilizer

Testing the sensitivity of photosynthetic parameters to climatic extremes

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Summary

The aim of the work was to characterize varietal differences and sensitivity of photosynthetic parameters, from the point of view of evaluating tolerance to drought and high temperature. To investigate this reliability, we tested a set of 10 pieces of the winter form of spring wheat (*Triticum sp.*) grown in pots under conditions of drought and high temperature. The non-invasive methods were used to identify the physiological properties of plants. The chlorophyll fluorescence fast phase parameters characterizing the structure and state of photosystem 2, were calculated based on measurements performed by Handy-PEA analyzer (Hansatech Instruments, GB), determination of relative water content (RWC), spectrophotometric determination of the content of free proline and assimilation pigments. As well as gasometric parameters characterizing overall photosynthesis, calculated from direct measurements of the infrared gas analyzer Licor-6400XT (Li-cor, Lincoln, NE, USA).

The problem of tolerance to environmental stress is very complex. The relative water content (RWC) in the leaves of individual genotypes exposed to dehydration and high temperature shows variability in values from 50 to 70 %, which results in differences in the regulation of the water regime under stress conditions. Based on the analyzes of fluorescence measurements, our results show that the influence of the stress factor was highly significant by reducing the maximum photochemical yield of PSII parameter (F_v/F_m), which led to inhibition the density of active reaction centers (RC/ABS) and a significant decrease in the total photosynthetic activity (PI_{abs}) in all genotypes, as well as the negative influence of the parameter W_k , characterizing the damage of the oxygen developing complex. The results also indicate stomatal closure and inhibition of carbon assimilation, which was manifested by stomatal inhibition of photosynthesis. Extensive experimental activities and approaches have allowed us to understand better the studied varieties' characteristics and responses to stressful conditions. The application of non-invasive measurements has proven to be a suitable method for direct use in cultivation practice.

Keywords: wheat, chlorophyll fluorescence, photosynthesis, drought, high temperature,

Physiological responses of drought-stressed soybean to exogenous hydrogen peroxide application

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Summary

The increasing population worldwide resulted in higher demand on food crops. However, non-favorable conditions, i.e., biotic and abiotic stresses caused serious yield losses according to several reports. More specifically, stress-sensitive crops were reported to have significant yield losses. Consequently, thorough work to select for stress tolerance and to find methods that can help plants overcome stress conditions was performed during the past decades and is still being practiced. Pre-stress exogenous application of hydrogen peroxide at low concentrations is gaining more importance as a cheap, easy-to-apply method for alleviating drought stress. An experiment was carried out in the Department of Applied Plant Biology at the University of Debrecen under hydroponic conditions in order to investigate the physiological response of a drought-sensitive soybean genotype “Coraline” to severe drought conditions applied during flowering stage, in addition to investigating the effects of exogenous application of 5 mM H₂O₂ on the drought-stressed plants. Total chlorophyll content, total carotenoids, potential and actual photochemical activity and relative water content were significantly reduced after 3 days of drought imposition. However, these traits were measurably elevated in plants pre-treated with H₂O₂ foliar spray. It could be concluded that pre-treatment with hydrogen peroxide might be an effective way of preparing soybean plants for an upcoming period of drought and, thus, better performing under such conditions. Future research will focus on the response to such conditions on the molecular level, along with engaging genotypes from different maturity groups to better understand whether different durations of life cycle have a role.

Keywords: abiotic stress, chlorophyll fluorescence, *Glycine max*, H₂O₂, photochemical activity

Effect of nitrogen fertilization on production and quality parameters of spring barley

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Summary

According to the EU plan for carbon neutrality, reductions for use of mineral fertilizer will be up to 20 % of current usage till 2030. Also, volatile prices of nitrogen fertilizers have a strong impact on farmers budget, which most often is connected with application of lower nitrogen rates. Therefore, the aim of this work was to investigate the influence of different amounts of nitrogen fertilizer in different locations in Croatia on the production and quality parameters of spring barley. A field trial was set up in 2022 growing season at three locations (Zagreb, Kutjevo, Osijek) as a part of long-term trial to investigate effect of four levels of nitrogen fertilization (0, 40, 80, 120 kg/ha) on yield, 1000-kernel, hectoliter mass. In addition to the production parameters of spring barley the basic quality parameters, starch, protein and fiber content were determined. Of the production parameters, different fertilization did not affect any of the tested parameters. On the other hand, the most important factor of variation was, as expected, the location. The location of spring barley sowing had a significant effect on barley yield, moisture, and 1000-grain values ($p \leq 0.001$). While the same value of 1000-kernels was the only one that differed statistically due to the combined influence of fertilization and location ($p \leq 0.05$). Qualitative parameters of spring barley grains differed statistically in the amount of fiber, and the more important qualitative parameter, starch, between locations and different fertilizations. Creating two groups of values with less fiber and more starch at fertilization of 0 and 40, respectively, more fiber and less starch at higher fertilization levels of 80 and 120 kg/ha ($p \leq 0.001$). We concluded, in short term experimental trial, that different nitrogen fertilization does not have that much influence on the production and quality parameters of spring barley, but the main variable in spring barley production is the location. That is, the results will in the future represent a good scientific basis in practical application for the production of spring barley with less application of nitrogen fertilizers to the soil.

Keywords: spring barley, fertilization, production, quality

Nitrogen deficiency reveals the importance of alternative electron flows in photoprotection of distinct wheat genotypes

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Summary

The most often limiting element for crop development and agricultural productivity is nitrogen, which is regarded as both the most significant and the main constraining factor. Our study aims to provide insight into the photosynthetic adaptation to various nitrogen availability by performing advanced analyses of CO₂ assimilation, PS II, and PS I photochemistry in wheat plants to investigate the effects of various nitrogen nutrition at three different leaf positions and assess the genotypic diversity using well-defined, distinct wheat genotypes (cv. Enola and cv. Slomer). Structural and functional changes associated with PS II photochemistry were analyzed by rapid chlorophyll fluorescence measurements recorded with a Handy PEA portable fluorimeter. Simultaneous measurement of PS I and PS II activity was performed with a Dual-PAM-100 system. Leaf photosynthesis was measured directly using an infrared gas analyzer, the Licor-6400XT. Using the Ciras 3 portable infrared analyzer, the method of rapid A/Ci curves was used to examine the photosynthetic potential and limitations. The measurements were performed separately on the youngest – flag leaf, the second row – of penultimate leaves, and the third row of leaves from the top. Our results demonstrated the decline in photosynthetic capacity, which was particularly pronounced in lower leaf positions. The modifications to PS II and PS I photochemistry, reflected non-stomatal limitations of photosynthesis. Low photosynthesis in N-deprived leaves, particularly in lower positions, was linked to a notable decline in the activity of alternative electron flows. An older genotype Slomer with a slower rate of CO₂ assimilation showed improved alternate electron flow and photorespiration capacity. In contrast, a modern, extremely productive genotype Enola responded to reduced photosynthetic activity by increasing nonphotochemical dissipation and cyclic electron transport. Our findings highlight how essential alternate electron fluxes are in eliminating the excitation pressure at the PS II acceptor side. A decline in linear electron transport resulted from the need to prevent the overreduction of the PS I acceptor side. The decrease in electron acceptor capacity was balanced by the structural and functional changes of the components of the electron transport chain.

Keywords: wheat, nitrogen, photosynthesis, photoprotection, photochemistry

Grain yield and quality of commercial oat cultivars in Croatia

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Summary

Oat products have recently gained increased attention in human consumption due to their favorable nutrient composition and water-soluble fiber components (β -glucan content). In addition, the physical traits such as kernel content and screenings are of prime importance in determining milling quality in husked oat. Thus, a main aim of this study was to determine the grain yield and quality of commercial oat cultivars currently grown in Croatia. A one-year field research was carried out in Gornja Posavina with five oat cultivars grown under recommended agronomic practices and operations. Oat grains were analyzed for a number of physical (1000-grain weight, test weight, groat percentage, screenings) and chemical (β -glucan, fat, protein) properties. As expected, tested cultivars significantly differed for grain yield and most grain quality traits. Cultivar Marta produced the highest grain yields and the largest test weight and 1000-grain weight. Cultivar Wiland had the smallest kernel (groat) content, while cultivar Baranja had the highest screenings (grains < 2.00 mm). Depending on cultivar, grain protein ranged from 11.3 to 12.8%, while fat content ranged from 5.07 to 7.72%. In contrast, relatively small cultivar differences were found for the β -glucan content (3.54 to 4.05%). Our findings highlight the importance of cultivar choice in the production of oat for the specific end-use production.

Keywords: β -glucan, groat content, protein, fat, screenings

The effect of biostimulators in conventional production on the yield and properties of wheat

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Summary

Since climate changes, reduced soil fertility and other stressful conditions have an increasingly negative effect on the success of wheat production, it is necessary to enable plants to alleviate stress. Biostimulators are physiologically active substances that promote the growth, development and nutrition of plants and help reduce the effects of stress caused by abiotic and biotic factors. The goal of the research was to examine the impact of biostimulators on the yield, agronomic and morphological properties of wheat compared to the conventional method of production on fertile soil. The field experiment was conducted during the 2021/2022 growing season in two treatments: basic control and control with biostimulator according in three repetitions. Weather conditions during the wheat growing season were mostly favorable because the total amount of precipitation was only 15 % lower, while the average air temperature was almost identical to the multi-year average. In the treatment with the biostimulator, the grain yield was higher around 7 %, the plant density around 6 %, the number of grains per spike around 3 % and the hectoliter weight around 2 % compared to the control treatment. A stronger positive effect of the biostimulator was most likely absent due to relatively favorable weather conditions during the wheat growing season and cultivation on soil with good fertility.

Keywords: biostimulator, wheat, yield, agroecological conditions, conventional production

BC maize hybrids yield in performance trials in 2022

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Summary

A total of 94 trials were processed in 2022 with an average of eight BC maize hybrids per location. It was the second consecutive year which was not favourable for maize production. During July, at the peak of maize fecundation, there was a period with long lasting drought followed by high temperatures, which continued during August. Such conditions were more pronounced in the eastern part of Croatia. The outcome of all processed results shows the average grain yield (AGY) of 8.74 t/ha, average grain moisture (AGM) at harvest of 18.51 % with an average plant density of 67.587 plants/ha. The analysis of the results shows the difference between maize yields from trials in Slavonia in relation to the yield from trials in western part of Croatia. This is a direct consequence of greater stress that maize suffered in the east of the country due to lack of precipitation. Thus, in four eastern counties BC323, a hybrid from FAO 300 achieved AGY of 8.08 t/ha with AGM at harvest of 15.93%, while in the other production counties it achieved AGY of 10,25 t/ha with AGM of 17,46%. Furthermore, one of the newest hybrids BC415 in the four most eastern counties had AGY of 7.64 t/ha with AGM of 17.04% and in other counties AGY 9.89 t/ha and AGM of 19.63%. This significant difference in grain yield and grain moisture is also visible in other hybrids: Agram, and Majstor. The best trial result in 2022 was obtained at the location Sudovčina (Varaždin county) with hybrid BC323 (16.19 t/ha).

Keywords: production trials, maize, hybrids, grain yield

Cyclic hydroxamic acid content of wheat (*Triticum aestivum* L.) under biotic stresses

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Summary

Cyclic hydroxamic acids are secondary metabolites of *Poaceae* species. These chemical compounds have diverse physiological roles. Stress metabolite function is their elemental task. The aim of the research was to establish the effect of a plant pathogen fungus and/or a pest on the cyclic hydroxamic acid content of two different wheat cultivars. At the age of three weeks, plants of two wheat cultivars, Hystar and KG Vitéz were treated. First group of plants was infested with *Fusarium graminearum* conidia, second with *Oulema melanopus* larvae, third with the previously mentioned fungus and insect together. The fourth group of plants made for control, which plants were not infested. Cyclic hydroxamic acid content was determined after 1 and 2 weeks of infestation. *Fusarium* inoculation significantly increased the Hystar hybrid's cyclic hydroxamic acid content compared to the control, from 104.34 mg kg⁻¹ fresh weight to 145.78 mg kg⁻¹ fresh weight respectively. On the other hand the cyclic hydroxamic acid content of KG Vitéz was significantly higher on time of second sampling compared to Hystar hybrid. The elevated level of cyclic hydroxamic acids support plants to overcome the negative effects of pathogens and pests. Plants with higher cyclic hydroxamic acid content can better prevent themselves from different biotic stressors.

Keywords: cyclic hydroxamic acids, wheat, plant resistance, biotic stress

Evaluation of green manures as a sustainable nutrient supply method in corn production

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Summary

The biggest challenges of future agricultural production are the activities against drought stress and the reduction of CO₂ footprint. Using of green manures can improve the soil water management, and it directly and indirectly contributes to reducing the ecological footprint of production. We set up our experiment in 2021 in Nyíregyháza, Hungary and investigated the forecrop effect of 4 different green manure plants with regard to the yield and yield parameters of corn on humic sandy soil. The experiment was set up in a randomized block design, the treatments were applied in 4 replicates, the size of one plot was 17 m². The examined green manure plant species were lupine, common vetch, oilseed radish and buckwheat compared to fertilization (80 kg ha⁻¹ N) and control treatments. Green manures were sown in August and were terminated in late October, corn was sown in the spring of the next year. The crop year was characterized by extreme drought, 44.5 mm of precipitation fell during the growing season of green manure crops, and 81.5 mm of precipitation fell during the growing season of corn. Based on the soil samples taken after sowing of corn, we found a more favorable soil moisture condition in the 0-50 cm layer of the soil in case of leguminous green manure forecrops compared to the control area. Based on the NDVI values measured by a handheld crop sensor (Trimble GreenSeeker) in corn, significantly higher values were observed in case of green-manured areas compared to the fertilized and control treatments. In case of yield parameters, based on the results of cob diameter and seed weight per cob, the values of the areas treated with lupine and common vetch significantly exceeded the values of the fertilized and control areas. Yield of the green-manured treatments achieved significantly better results than the fertilized and control areas (2.6 times more yield in case of lupine and common vetch forecrop, 1.4 times more yield in case of oilseed radish and buckwheat forecrop). Based on our results, despite the drought situation, leguminous green manures with low water consumption function as an excellent nutrient supply alternative in corn production.

Keywords: green manure, corn, soil moisture, yield, NDVI

The influence of conservation tillage systems on soybean weediness

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Summary

Conservation tillage systems, compared to conventional ones, have proven to be more effective from a biological, ecological and economic point of view, while at the same time preserving the ecosystem. Research with conservation tillage systems was conducted in 2022 on the experimental area of Križevci College of Agriculture (46° 01' 38" N, 46° 33' 32" E). The field experiment was set up according to a split plot experimental design with three main tillage treatments: ST (conventional, plowing), CTD (conservation, loosening with a minimum of 30% crop residues on the surface) and CTS (conservation, tillage up to 10 cm with a minimum of 50 % crop residues on the surface). Weed sampling was carried out during the critical period for weediness in soybean crops. The number and above-ground biomass of weeds was determined by counting individual weed species using a square of 0.25 m² in four replicates per experimental plot. Weeds from each square were cut at ground level, counted, sorted according to species, dried at 65 °C and weighed. The applied herbicide treatments were uniform on all tillage treatments. Tillage had a statistically significant effect on the total number of weeds, the above-ground biomass and the number of weed species. The highest number of weeds was found in the CTD treatment (44.67 m⁻²), and the lowest in the ST treatment (7.33 m⁻²), with a statistically significant difference between ST and conservation treatment treatments. The CTS treatment resulted in the highest weed biomass (48.77 g m⁻²) but without a statistically significant difference compared to CTD (39.98 g m⁻²). The highest number of weed species was found in the CTS treatment (3.66 m⁻²), and the lowest in the ST (1.33 m⁻²), with statistically significant differences between all tillage treatments. All investigated weed parameters were, on average, the lowest in the conventional tillage system compared to the conservation systems. The CTD treatment resulted in a smaller number of weed species and a lower biomass compared to the CTS treatment, which indicates the need for further research into the impact of conservation tillage on the occurrence of weeds in soybeans.

Keywords: conservation tillage, weeds, soybeans

This research work has been financed by the Croatian Science Foundation under the project "Assessment of conservation soil tillage as advanced method for crop production and prevention of soil degradation – ACTIVEsoil" (IP-2020-02-2647)

Frequency distribution of Nitrogen balances in Hungarian crop production (2016-2018)

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Summary

For the management of crop macronutrients, especially in the cases of nitrogen (N) and phosphorous (P), farmers should take into consideration both agronomic and agro-environmental aspects. Excess of N (and P) application above crop demands can be an environmental threat for ground- and surface waters which must be avoided. In 1991, the EU introduced the Nitrates Directive (91/676/EK), which aimed to reduce water pollution caused or induced by nitrates from agricultural sources. The Directive requires Member States to apply agricultural action program measures throughout their whole territory or within nitrate vulnerable zones (NVZs) (Hungarian Government Decree 27./2006./II.7./), requiring to promote best practice in the use and storage of fertilizer and manure on these special areas (FVM Decree 59./2008./IV.29./). Evaluating the national nitrate database for 2016 to 2018, focusing on the five major crops (rapes, barley, sunflower, winter wheat and maize), resulted in receiving more accurate data on the yearly and three-years accumulated agro-environmental nitrogen balances. Frequency distribution of N balances of the five main crops showed a similar shape, i.e. Gaussian curves. The highest N balance frequency distribution curve maximums were found in the case of sunflower and rapes in the investigated years ranged -10 to +20 kg ha⁻¹. The lowest N balance frequency distribution curve maximums were recorded under maize crop in each of the three years ranged -90 to -70 kg ha⁻¹. N balance frequency distribution curve maximums of barley and winter wheat were in between of the five main crops ranged -50 to -30 kg ha⁻¹.

Keywords: nitrates database, Gaussian curves, five main crops

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Acetic acid as drought alleviator in different genotypes of sweet corn

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Summary

In crops, it has recently been reported that the application of acetic acid at low concentrations reduces the effects of drought. The current study was to seek the effective mitigation measure of PEG-induced water deprivation under hydroponic conditions for different genotypes of sweet corn (Dessert, Tyson, Messenger, Royalty, and Gss 8529/M) by exploring the potential role of acetic acid. Sweet corn seedlings (V5-V6 stage) were grown in 5% PEG solution until they went through drought-like symptoms (visible/physiological). Twenty-five-day-old (V7-V8 stage) plants were treated with two different concentrations of acetic acid (20mM and 10mM) via the foliar application for five consecutive days. For control plants, distilled water was applied. Photosynthetic pigments and stomatal conductance in different genotypes showed variable responses towards water deprivation and acetic acid treatments. Stomatal conductance decreased in four genotypes except for Royalty under water deprivation compared to control. A significant decrease in stomatal conductance was recorded in the Gss 8529/M genotype when treated with 20mM acetic acid + PEG compared to control and PEG treatment. However, Dessert recorded a significant increase in stomatal conductance when treated with 10mM acetic acid under water deprivation. Compared to the control, chlorophyll-a and chlorophyll-b increased in all genotypes in the PEG treatment. In the Royalty genotype, a significant decrease in total carotenoid content was recorded in both acetic acid treatments compared to water deprivation. In Tyson, a significant decrease was recorded under 20mM acetic acid + 5% PEG compared to PEG treatment. It is concluded that the effect of water deprivation on photosynthetic pigments in sweet corn varies from genotype to genotype. The role of acetic acid of this concentration on photosynthetic pigments under water deprivation in sweet corn at the V7-V8 stage did not show promising results, however, needs further research to analyse various morphological, physiological, biochemical and molecular responses of these genotypes at different growth stages, concentrations of drought and acetic acid.

Keywords: water deprivation, acetic acid, drought alleviation, chlorophylls, sweet corn

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Influence of intercropping maize with climbing bean on forage yield and quality

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Summary

Maize forage is poor in protein content which shows its low quality and nutritive value. Regarding to high feed costs of protein supplementations, legumes can be used in livestock nutrition for their high protein content and, thus, providing cost savings. Since legumes have low dry matter yield, acceptable forage yield and quality can be obtained from intercropping cereals and legumes compared with their sole crops. Maize (*Zea mays* L.) and climbing bean (*Phaseolus vulgaris* L.) intercropped in different sowing densities and pure maize crops were evaluated to the best intercropping system with respect to forage yield and quality. Maize was cultivated alone (75 000 plants ha⁻¹) and intercropped with climbing bean as follows: 75 000 plants ha⁻¹ of maize and 37 500 plants ha⁻¹ of bean (MB₁), 75 000 plants ha⁻¹ of maize and 50 000 plants ha⁻¹ of bean (MB₂) and 75 000 plants ha⁻¹ of maize and 75 000 plants ha⁻¹ of bean (MB₃), in rows alternating with maize. The highest dry matter yield was produced by MB₃ (21.9 t ha⁻¹), and the lowest by solo maize (18.3 t ha⁻¹). All intercrop had higher crude protein values in dry matter 96 g kg⁻¹ for the MB₁, 106 g kg⁻¹ for the MB₂ and 110 g kg⁻¹ for the MB₃ than the monocrop maize (76 g kg⁻¹). Intercropping of maize with climbing bean reduced neutral and acid detergent fiber content, which in turn, resulting in increased forage digestibility. Based on forage yield and quality, this study showed that among all intercropped forages the MB₃ treatment was better performing than other intercrops.

Keywords: intercropping, maize, climbing bean, yield, quality

Industrial hemp yield components and stem chemical parameters in relation to presowing potassium application

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Summary

Industrial hemp is grown mostly for its seeds and fiber. The aim of this study was to determine the industrial hemp yield components and elements in stem with pre-sowing potassium fertilization of different formulations: K0=control, K1=100 kg/ha KCl and K2=100 kg/ha K₂SO₄. Monthly amounts of precipitation and mean air – temperatures were monitored. In this study several yield components were determined: the number of plants per square meter, stem height, seed mass (grams per plant), and finally, industrial hemp seed yield per unit area. After harvesting and collecting of the plants, the average number of plants per square meter were 32, the average stem height was 71.62 cm, while the average seed mass (grams per plant) was 1.814 g. Highly significant correlation was found between the mass of seeds per plant (g) and the height of the plant $r=0.320$. According to the regression analysis in this research, it was determined that there is a very weak connection between the height of the stem and the mass of the seeds, while it was determined that for each centimeter of increase in the height of the stem, and the mass of the seeds increases only by 0.02 g per plant. Average content of potassium in the stem was between 1.15% (K0) to 1.39% (K2). The average yield of industrial hemp seeds was 0.43 t ha⁻¹.

Keywords: industrial hemp, potassium, plant height, stem, yield components

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Tradition of industrial hemp production in Croatia

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Summary

The production of industrial hemp was primary for fiber extraction. In Croatia in the first half of the 20th century approximately occupied between 8,000 and 12,000 hectares. The largest sown areas were in 1949 when industrial hemp was grown on about 21,000 ha. In the 1960s, industrial hemp was grown on an average of 7,165 ha, with an average yield of dry stem 5.81 t ha⁻¹ and 0.88 t ha⁻¹ of fiber. In the next decade (1970-79) areas were reduced to 2,331 ha and even less, in 1980s harvested area reduced to 1,131 ha. Even the areas was reduced, the stem yield increase up to 8.79 t ha⁻¹ (1980-89). In the 1990s production of industrial hemp for fiber extraction in Croatia was gradually abandoned. According to statistical data, the last areas under industrial hemp intended for fiber extraction were in 1995 on only 30 hectares. In Slavonia and Baranja in the first half of the 20th century larger hemp factories were in Vukovar, Vladislavci, Osijek and Darda and in the second half of the 20th century in Viškovci and Črnkovci. The factory in Črnkovci was active for the longest time (until 90's). Revitalization of industrial hemp production in Croatia began in 2012, when is primarily grown because of the seeds from which the oil is extracted. By changing the law in Croatia, from 2019 it is allowed to use the whole plant, which will certainly lead to the development of new products and expand the interest of farms in the production and processing of industrial hemp.

Keywords: industrial hemp, processing, Croatia, production, fiber

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Evaluation of yield components of white lupine under different ecological conditions

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Summary

Lupine (*Lupinus albus* L.) can be used in human nutrition because of its high protein and oil content and as a green manure, as it contributes to improving soil structure, increasing organic matter content and accumulating nitrogen and phosphorus, which is beneficial on nutrient-poor soils. It also has a place as a green fodder and as a protein supplement for animal feeding. Most European plant protein requirements are covered by imports, this model of agricultural production can lead some environmental problems, therefore increasing the cultivation of legumes in Europe could be a promising alternative. In order to ensure a stable seed stock of these alternative plant species, it is necessary to adapt the cultivation technology to different ecological areas.

In 2021, lupines were sown in four agroecological conditions in Hungary with different soil properties in order to determine the factors influencing the evolution of the yield and yield components. We applied different row spacings, 75 cm, 24 cm and 12 cm. During the experiment, we examined soil penetration, total biomass weight, plant height, number of pod levels, number of pods per levels, number of seeds per pod, the number of seed and seed weight per plant and thousand seed weight. Based on the results among the four locations, we experienced favorable development in the areas where 75 cm row spacing were used and soil pH values were acidic. The yields varied between 610 and 1158 kg ha⁻¹. The proportion of the vegetative mass compared to the seed weight varied between 70 and 73% in the investigated fields.

Keywords: white lupine, row spacing, soil pH, pod number, seed yield



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The state of the European eel, *Anguilla anguilla* (Linnaeus, 1758) population in the Neretva delta area

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Summary

In the area of the Neretva delta, where the European eel, *Anguilla anguilla* (Linnaeus, 1758), has been traditionally fished for centuries, as well as in most parts of Europe, a decline in its population was noticed. Given that regular monitoring is not carried out in Croatia, nor have minimum fishing sizes been determined, the goal of this research was to obtain detailed data that will serve as a “zero state” for the adoption of future management plans. Eel samples were collected during joint field work with fishermen as part of the project “Fisheries and Science Network of the City of Ploče” within the framework of Measure I.3. “Partnership between scientists and fishermen for the period 2017-2020 “. Fyke net and eel traps with a net weight of 12 mm, were used for fishing in two localities: Neretva delta and Mala Neretva. Joint field activities with fishermen were carried out during January, March, May, June, July, August, September, and November 2021. A detailed morphometric analysis was performed on the samples, which included determining the life stage of the eel (yellow or silver eel). Length-weight ratios, Fulton’s condition index, ratio of maximum height to total length, hepatosomatic index and index of visceral fat tissue were determined. Features of the digestive system and air bladder were also analyzed in detail. In addition to the mentioned parameters, at each field trip, the amount of catch in the traps was recorded, and based on this data and discussions with the fishermen, an estimate of the average monthly eel catch per one trap was achieved. In addition to the above, a health examination was carried out, which included a microbiological examination, examination of skin and gill swabs using the eDNA analysis method, and parasitological and virological examination for the presence of EVEX and AngHV-1. To determine the possible influence of toxic substances, the concentration of mercury and 14 organochlorine pesticides in the meat of each individual was analyzed. To increase the reliability of the results for the parameters for which seasonality was monitored, the samples were grouped by seasons and stations whenever this was possible considering the number of samples in the group. The results show that numerous measured parameters can affect the size of the population, but with proper management measures it is possible to increase its population size.

Keywords: eel monitoring, management plan, population size, yellow and silver eel

Aquavoltaics - floating solar panels in synergy with aquaculture

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Summary

Solar photovoltaic (PV) technology is the most widely accessible sustainable and clean source of energy that can be scaled to meet humanity's energy needs. Floatovoltaic (FPV) represent an emerging power-generation technology utilizing idle water and solar energy systems - comprising floating photovoltaic panels over water. In terms of the environment and other variables, it is possible to install various forms of FPV on water surfaces, such as reservoirs used for irrigation purposes - motivated by the increasing demand for energy in modern irrigation systems and agriculture. Other potential applications include quarry and natural lakes, wastewater tanks, oceans and fish ponds - integrated into aquacultures as aquavoltaics (AquaPV). The goal of AquaPV is the efficient use of water with the dual benefit of food and energy production. The cultured species have different requirements, confirming that the essential parameters must vary depending on the species and the husbandry system. The AquaPV approach aims to maintain parameters such as water and air temperature, light availability, water pH, dissolved oxygen, feeding system, and predator pressure, and improve the system by exploiting synergies between aquaculture and FPV systems. There is limited knowledge of the effects of FPV on the receiving water body. The addition of floating modules will most likely complicate aquaculture system maintenance, and aquatic life may also slow or disrupt PV module maintenance. To avoid the increasing consumption of land, the approach of aquavoltaics offers a solution for the dual use of land. In addition to the advantageous shading for aquatic animals in summer, the strong reduction of water loss due to lower evaporation rates is particularly interesting.

Aquavoltaics is a new concept that combines two areas where significant research is needed. Predicting changes in water body processes, characteristics, and services as a result of FPV implementation is a critical knowledge gap that can lead to poor societal decisions and poor management of water bodies. With a suitable system approach, AquaPV can contribute to sustainable water use and fulfil the concept of the food-water-energy nexus.

Keywords: aquavoltaics, floating photovoltaic, aquaculture, sustainable, water

Influence of abiotic factors on the growth of the Mediterranean mussel *Mytilus galloprovincialis* nearby cage farming of European seabass

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Summary

This study presents the results of measuring morphological characteristics of the Mediterranean mussel *Mytilus galloprovincialis* at two locations, near the sites of European bass farming. The measurements were carried out 6 months after settlement of mussels on collectors in Lake Prokljan and in the Middle Channel – the central eastern Adriatic Sea. The difference between the two locations is due to abiotic factors, mainly salinity. The aim of this research was to determine the influence of abiotic factors on mussel settlement and growth, and the influence of intensive fish farming on mussel growth. Therefore, the morphological characteristics (length, height, width and weight) of mussels were measured at two locations, near fish cages. The average measured length, width, and weight (total weight and wet tissue weight) of mussels after 6 months were higher in mussels from Lake Prokljan, while the average height and shell weight were higher in mussels from the Middle Channel. The results show faster growth of mussels in Prokljan Lake which is most likely due to the influx of low-salinity surface water (6.6 ‰). In addition, a higher settlement was recorded at the collectors in Prokljan Lake, which is most likely due to the fact that this location is less exposed to currents and waves throughout the year compared to the Middle Channel.

Keywords: *Mytilus galloprovincialis*, integrated aquaculture, abiotic factors

Metric and spectral insight into bee pollen-to-bee bread transformation process

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Summary

Due to numerous bioactive constituents, both bee pollen and bread represent a potential functional food. The conversion of bee pollen into bee bread is a complex biochemical in-hive process that enables the preservation of the pollen's nutritional value. The aim of this study was to determine the depth of the honeycomb cells to which bees store pollen and to provide a spectral insight into the chemical changes in pollen composition from the initial raw material (bee pollen) to the final product (bee bread). The study was carried out on three experimental colonies of Carniolan honey bee (*Apis mellifera carnica*) from which fresh bee pollen was collected using front-mounted pollen traps, while the bee bread samples were manually extracted directly from the honeycomb cells. Along with sampling, the depth of empty honeycomb cells and cells with stored pollen content was measured using a caliper. Bee pollen and bee bread samples were analyzed by Fourier transform infrared spectroscopy (FTIR) coupled with a single-reflection diamond ATR (attenuated total reflectance) accessory. The results showed that the average depth of empty cells was 11 mm, and that the bees stored bee pollen up to an average of 7.85 mm of cell depth thus covering between $\frac{2}{3}$ and $\frac{3}{4}$ of the honeycomb cell. The FTIR-ATR analysis revealed a unique IR spectral pattern of both the bee pollen and bee bread samples, indicating compositional changes primarily reflected in higher water content and altered composition of the carbohydrate fraction (to a lesser extent the lipid fraction) in the bee bread compared to the bee pollen.

Keywords: bee pollen, bee bread, bee pollen-to-bee bread transformation process, honeycomb cell depth, FTIR-ATR spectroscopy

Selective breeding approach to improve feed efficiency in African catfish (*Clarias gariepinus*)

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Summary

A selection programme was conducted on African catfish, an important species in intensive aquaculture, to improve utilisation of fishmeal poor feeds. Offspring were produced with 4 multifactorial crosses of 5 males and 5 females in each generation, and body measurements were performed at market size (N=11258). After applying a selection pressure of 20% to body mass in flow-through systems on a semi-industrial scale, the effect of selection was tested in both flow-through and recirculating aquaculture systems (RAS) after two and three years of selection, respectively. In the flow-through system, the direct selection gain for body mass was 11% and 23% in generations F3 and F4 fed the experimental diet and 14% and 26% when fed the control diet, suggesting a relevant commercial potential of artificial selection in this species. However, no feed-specific selection gain was detected in the flow-through system, which could be explained by the high selection response in the groups fed the control diet. In RAS, the average selection gain when fed the experimental diet was 32% and 33% and when fed the control diet was 12% and 12% in generations F3 and F4, respectively. In this system, there was also a feed-specific selection gain of 21% in both generations. The estimated heritability for body mass (0.33-0.60) was similar in the different systems and the feed conversion ratios for the different feeds were almost the same in RAS.

The work was supported by the by iFishIENCi project (European Union's Horizon 2020 research and innovation program under grant agreement No 818036), by the National Research Development and Innovation Office (NKFIH) Hungary, grant number 2017-2.3.3-TÉT-VN-2017-00004, and the EFOP-3.6.3-VEKOP-16-2017-00008 project, which is co-financed by the European Union and the European Social Fund.

Keywords: African catfish, *Clarias gariepinus*, low fish-meal feed, selective breeding

The potential use of biodegradable materials in the Adriatic Sea small scale longline fishery

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Summary

Longlines are a widely used fishing gear. The longline consists of a mainline with a series of hooks attached to thinner monofilament snood lines usually made of nondegradable plastic material such as nylon. During longline fishing, some of the snood lines are lost at sea contributing to plastic pollution in the oceans. The risk for the snood line loss is often unknown and can vary between different longline fisheries and fishing grounds. Replacing nylon with biodegradable plastics could reduce the plastic pollution caused by this fishing gear. In this study, we estimated and compared the risk for the snood loss and catch composition in the small-scale coastal longline fishery of the Adriatic Sea. We also compared catch efficiency between biodegradable and nylon snood lines for capture of two-banded sea bream (*Diplodus vulgaris*), common pandora (*Pagellus erythrinus*) and axillary sea bream (*Pagellus acarne*). No significant differences were found between the two materials. Therefore, this study shows that the use of biodegradable plastic materials has the potential to reduce fishery related plastic pollution without affecting the catch efficiency of the fish species studied.

Keywords: marine pollution, microplastic pollution, longline fishery, biodegradable materials, catch comparison

Other records of *Melibe viridis* (Kelaart, 1858) in the southeastern Adriatic Sea

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Summary

Melibe viridis (Kelaart, 1858) is a nudibranch native to the waters of the western Indo-Pacific. It was first recorded in the Mediterranean Sea in 1970, where it presumably entered through the Suez Canal. The first record of this species in the Adriatic was in October 2001 in the bay of Stari Grad on the island of Hvar. After that, its occurrence was reported in 2004 near the town of Herceg Novi (Montenegro). Reproductive specimens were detected in October 2014 in the bay of Boka Kotorska (Montenegro), where two adults were found spawning at a depth of three meters. In this paper, further records in the southeastern Adriatic Sea are described. The study area is located in Bistrina Bay, a part of Mali Ston Bay, which is an area protected at the national level as a Special marine reserve. The hydrological and ecological conditions in Bistrina Bay depend largely on the influences of the mainland and the strong underwater freshwater springs in the inner part of the bay. In addition to the high biodiversity, this area has been important for shellfish farming since Roman times. Currently, Mali Ston Bay has the highest production of the European flat oyster, *Ostrea edulis* in the Mediterranean.

M. viridis was first sighted in Bistrina Bay on 11 November 2021, with two specimens crawling near to the freshwater spring below the shore. On November 15, two 50-meter belt transects were established parallel to the shoreline. The transects were placed five meters from the shore at a depth of four meters on a sandy, muddy bottom. The number of specimens and their spawn was noted by a diver along each transect and recorded on video covering a 50 x 2 m area. A total of 20 specimens and 22 egg ribbons were recorded in both transects, indicating massive spawning. The average length of the specimens recorded during this survey was 17.75 ± 5.46 cm, with the largest specimen measuring 27.00 cm and the smallest 10.00 cm. The finding that this species continues to expand and establish viable populations throughout the Adriatic Sea highlights the need for continued monitoring of areas that provide important ecosystem services.

Keywords: *Melibe viridis*, Bistrina Bay, new records, southeastern Adriatic Sea

Activity pattern of European mouflon in the Mediterranean habitat - a preliminary results

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Summary

Animals allocate their time between activity and rest, with the trade-off reflecting the need to satisfy basic requirements while minimising energy costs. Activity level, the proportion of time spent actively, serves as a key measure for understanding such a trade-off. Activity levels can be affected by several factors, including population density, resource availability, climate, predation risk and mating season, which often occur synergistically, thus making it difficult to distinguish the effects of each factor. We quantified the activity level of European Mouflon in two different habitats, Senj and Dugi Otok areas. Large predators (grey wolf, Eurasian lynx and brown bear) are present in the Senj area, while no large predators occur on the island of Dugi Otok. Cfb climate prevails in the Senj area, while Cfa climate prevails in the Dugi Otok area. On the island of Dugi Otok and in the area of Senj, 27 and 22 camera traps were placed, respectively, in a systematic grid of 1.5 km x 1.5 km. The cameras were active between April and June 2022. Photos were sorted and processed in Agouti software to obtain mouflon occurrence frequency. Output datasets were processed in R using the “camtraptor” and “activity” packages to calculate and plot activity levels. On the island of Dugi Otok, a bimodal activity pattern was detected with activity peaks at dusk and dawn. In the Senj area, a trimodal activity pattern was detected with activity peaks at dusk, midday and dawn. Nocturnal activity was detected on the island of Dugi Otok, while no nocturnal activity was detected in the Senj area. While several factors influence activity patterns, we assume that the trimodal activity pattern is mostly a result of population density and predator activity. The nocturnal activity detected on the island of Dugi Otok is mostly due to the absence of large predators.

Keywords: Adriatic Sea, camera trap, locomotor activity, *Ovis gmelini musimon*, ungulate

The possibility of commercialization of the catch of the invasive Atlantic blue crab, *Callinectes sapidus* (Rathbun, 1896)

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Summary

The Atlantic blue crab, *Callinectes sapidus* (Rathbun, 1896), is native to the western Atlantic and is distributed from Nova Scotia, Maine, and northern Massachusetts to Argentina. It has also been accidentally or intentionally introduced into Asia and Europe, which is most often attributed to the introduction of larvae via ship ballast. The first finding of a blue crab on the Croatian side of the Adriatic dates back to 2004. Later, more specimens were found in the wider area of the Neretva river estuary. Research on the position of the blue crab in the food chain of the Neretva Delta showed that the blue crab is in competition with native species. Due to its agility in movement and high fecundity, it spreads rapidly to the surrounding areas, which represents a threat to local biodiversity. For the fishermen, the blue crabs they pulled out of the water, entangled in torn nets, were a real disaster and an unsolvable puzzle. With their sharp carapace and claws, they destroy fishing nets, and feed on fish caught in nets or traps. But there is another, opposite side to the story of this species. Blue crab is the fifth most popular crab on the world market. It is especially in demand on the Asian, American, and Australian markets, where it is a delicacy on the menus of many restaurants. Therefore, there is a possibility that this pest, as we see it today, can be turned into a significant commercial catch, which would automatically reduce its population or keep it under control. Such an approach brought good results in Greece and Tunisia, where fishermen have been trained for blue crab fishing, and restaurants have introduced known recipes for their preparation and developed new ones. The paper considers and proposes activities of a similar approach to the commercialization of blue crab on the Croatian coast, while referring to existing activities that can be considered as the beginning of this process.

Keywords: *C. sapidus*, blue crab commercialization, gastronomy, market

Controlled spawning of grass carp (*Ctenopharyngodon idella*) and common carp (*Cyprinus carpio*) in the Crna Mlaka fish farm

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Summary

Controlled spawning facilitates manipulation of broodstock and larvae and allows easier monitoring of the production process itself. In addition to aquaculture, the Crna Mlaka fish farm is also distinguished by the fact that it is a special ornithological reserve, declared in 1980 and listed in the Ramsar Convention since 1993. Controlled spawning of grass carp (*Ctenopharyngodon idella*) and common carp (*Cyprinus carpio*) was carried out in Crna Mlaka fish farm from April to June 2022. The dry method of controlled spawning was used, followed by incubation of common carp eggs in McDonalds containers. After hatching, the larvae were transferred to Weiss's containers. Grass carp eggs were transferred directly into Weiss's containers after fertilisation. From common carp eggs incubated at 22 °C, larvae hatched after 85 hours. Grass carp eggs hatched after 25 hours. In the study, the average body weight of female common carp broodstock was 7.47 ± 1.68 kg, while they produced 0.875 ± 0.329 kg of fry. Grass carp female broodstocks had an average body weight of 9.82 ± 0.98 kg, and they produced 0.89 ± 0.195 kg of fry. The average value of the ratio of eggs produced to body mass was 11.97% for common carp and 9.27% for grass carp. In general, controlled spawning results in better spawning success and a higher total amount of larvae produced in freshwater aquaculture.

Keywords: grass carp, common carp, controlled spawning, Crna Mlaka

Seasonal variability and repeatability of some Varroa resistance traits

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Summary

The ectoparasitic mite *Varroa destructor* is nowadays considered the greatest threat to the health and survival of honey bee colonies (*Apis mellifera*). As chemical treatment of honey bee colonies becomes less effective against the Varroa mite, new solutions must be found to protect honey bees from this pest. Breeding bees resistant to Varroa is often considered a long-term and sustainable solution. In this study, we measured the seasonal variability and repeatability of suppressed mite reproduction (SMR) and brood cell recapping. Brood samples were taken six times during the season (May to September) from 52 honey bee colonies. In total, 158 brood samples were included in statistical analysis, in which at least 10 brood cells were infested with a single foundress Varroa mite. A total of 4040 infested brood cells were examined, averaging 25.57 infested cells per sample. Repeatability was calculated using a sample size of 34 colonies, of which three to six brood samples with at least 10 single infested brood cells were from one colony. The repeatability value can be used as an upper limit of heritability or to show what the gain will be for repeated measures. There was a significant difference in the recapping of infested brood cells between sampling months, while there was no difference in the proportion of non-reproducing mites. The calculated repeatability for recapping infested cells was 0.391 and for SMR was 0.353.

Keywords: *Apis mellifera*, resistance traits, *Varroa destructor*

Analysis of land structure in non-hunting areas

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Summary

In the Republic of Croatia, a total of 1,084 hunting grounds have been established as open hunting grounds, fenced hunting grounds and game breeding grounds, which, according to the data of the Croatian Bureau of Statistics, cover 4,700,000 ha of hunting and 700,000 ha of non-hunting areas, i.e. a total of 5,400,000 ha, which is 95.42 % of the total land area of the Republic of Croatia (5,659,400 ha). The establishment of hunting grounds is prohibited in the areas where hunting is prohibited by the law that establishes its purpose, such as construction areas (settlements), public roads and other public lands, and these areas are not fenced. Therefore, various wildlife species pass through or emerge in these areas, and occasionally they permanently inhabit these areas in a certain number, although they are not their habitat. The protection and hunting of wildlife in these areas is regulated by the Wildlife Protection Program, which defines the number of wildlife that are permanently in these areas and are occasionally or temporarily retained. For this research, data were collected from 279 established wildlife protection programs on the land structure totaling 362,082 ha or 51.73% of the non-hunttable area. Data were collected on the land structure in the administrative boundaries of 75 cities for 204,929 ha (56.60%) and 204 municipalities for 157,153 ha (43.40%). The proportions of land in relation to cities and municipalities are 43.03% for building land versus 58.51%, 9.10% for forest land versus 6.09%, 39.86% for agricultural land versus 25.65%, and 8.01% for other public areas versus 9.75%. It is noticeable that agricultural land is disproportionately represented in the land structure of cities with 81,692 ha in cities and 40,150 ha in municipalities, as well as forest land with 18,645 ha in cities and 9,565 ha in municipalities. Rural areas municipalities have larger areas of building land with 91,954 ha compared to 88,171 ha in cities. The obtained data open space for further analysis of the land structure of non-hunting areas and monitoring of game occurrence on them and in settlements.

Keywords: non-hunting areas, wildlife, Wildlife Protection Program, cities, municipalities

Seasonal variations in the structure and abundance of the mesozooplankton community in the port of Rijeka

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Summary

This survey aimed to update the mesozooplankton community's existing database in the port of Rijeka. The port baseline survey protocol developed during the BALMAS project (2013 - 2016) served as the guideline for the sampling methodology of this study. The port of Rijeka is located in the northern Adriatic Sea, its border is 100 m isobath. The northern Adriatic Sea is characterized by the largest riverine contributions in the basin. The area of the Port of Rijeka is enclosed and has a connection with the open sea in the western part which is quite narrow. Weak to moderate easterly winds affect the port, blowing the water out to the open sea. Average abundance values of total mesozooplankton ranged from a minimum of 2,888 ind. m⁻³ at station RIbwB during the autumn period to a maximum of 99,365 ind. m⁻³ at the same station RIbwB during the summer period. Copepods were the most abundant group throughout the study period, dominating all samples. The minimum contribution of copepods to total zooplankton was 56% in autumn at station RIbwB, while the maximum contribution of copepods was 96 % in spring at station JPG28. Values of temperature, salinity, and total net zooplankton abundance were correlated with individual zooplankton groups in each season. Correlations between important zooplankton groups during each season were also performed. The highest number of significant correlations of temperature, salinity, and total net zooplankton with individual zooplankton groups was observed in spring and, the highest number of significant correlations between groups was observed in winter and spring. The average abundance of total zooplankton and copepods increased compared to previous research conducted in the same area in 2014.

Keywords: mesozooplankton, port of Rijeka, copepods, community structure, abundance

Evidence of hybridization between mouflon and domestic sheep

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Summary

Hybridization between related species plays an important role in the adaptation and evolution of ungulate species. The European mouflon is an ungulate species that has been introduced into the Mediterranean habitat and often shares its habitat with traditionally kept domestic sheep. Since the two species are closely related, there is an increased chance of hybridization in shared habitats. In this study, we analyzed a possible hybridization between European mouflon and domestic sheep on Dugi Otok island. Using ovine medium-density SNP array, we genotyped 21 mouflon, 15 feral domestic sheep, and two individuals identified as potential hybrids for their phenotypic traits. The ADMIXTURE analysis was able to identify two animals as hybrids in which 0.12 and 0.26 genomes of domestic sheep introgressed into the mouflon genotype, respectively.

Keywords: hybridization, mouflon, domestic sheep, SNP

Occurrence of microplastics in dried fish

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Summary

Dried fish is considered as a prominent and affordable low-cost protein source for people in developing countries. Fish used as raw material for processing is often contaminated with microplastics (MPs) in the aquatic environment. Furthermore, the unsanitary processing of dried fish products often results in presence of unwanted particles such as metals, microplastics etc., which endanger the health of consumers. In the present study, samples of the two most common carnivorous fish species spotted snakehead, *Channa punctata* (freshwater species) and Bombay duck, *Harpadon nehereus* (marine species) were collected from Jagiroad Dry Fish Market, the largest dry fish market in India, and analysed for the presence of microplastics motes. MPs were extracted with cellulose filter paper from the whole fish body (n=10 per species) using KOH and NaI digestion, and examined physically under the microscope, while their chemical composition was determined by FTIR. An average of 51 and 42 MP motes were found per individual of *C. punctata* and *H. nehereus*, respectively. In both fishes, MPs predominated in the form of fragments. High-density polyethylene (HDPE) was the predominant fraction in freshwater dry fish, while polyvinyl chloride (PVC) was more prevalent in marine fish. Dried fish is usually consumed whole or used in the preparation of fish feed. In both cases, it represents a direct or indirect potential health risk. Therefore, further research is needed to identify the source of these MPs, and to develop new methods for its removal from dried fish products.

Keywords: microplastics, dried fish, fish products, contamination, health risk

Overview of fish farming cage structures and their applicability for offshore mariculture in the Adriatic Sea

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Summary

In order to continue the development of mariculture in a sustainable manner, fish farms tend to be relocated to further offshore sea areas. In recent decades, a large number of offshore cage farms have been designed, built, and put into use, which only implicates further development in this direction. An overview of existing mariculture structures was conducted to analyse the technical characteristics of various cage designs and their applicability to potential offshore farm locations in the Adriatic Sea. An extended SWOT analysis was conducted to single out systems with greater application potential, taking into account conditions in the Adriatic Sea and current fish farming practices. Various options are ranked and a preliminary solution of the regionally applicable fish farming cage is proposed to allow further refinement of the conceptual designs taking into account not only the technical features but also other factors. This study discusses technical parameters that must be considered for effective offshore mariculture, in addition to being combined with other parameters to fully support production.

Keywords: mariculture, offshore, rearing cages, technical solutions, Adriatic Sea

Consumer attitudes, preferences, and perceptions regarding the consumption of game meat

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Summary

Game meat or venison, is the meat of killed large or small game intended for human consumption. Exceptionally, it may also be obtained by slaughtering certain species of intensively farmed game. The consumption of game meat in Croatia is significantly lower than that of meat from domestic animals. The aim of the paper is to determine the attitudes, preferences, and perceptions of consumers regarding the consumption of game meat in Croatia, by means of a questionnaire. A total of 845 respondents from all over Croatia were surveyed, of which 715 were hunters. Although game meat is available to most respondents (67.92%) on a daily basis, most (40.47%) consume it only occasionally and only a few (7.81%) once a week with meat products predominating, the most common being game salami (62.25%). Wild boar meat (33.42%) is the most consumed followed by rabbit (19.93%), pheasant (16.47%) and venison (13.56%), while other types of game meat are consumed by less than 7% of respondents in total. The majority of respondents (78.82%) were aware of the nutritional value of game meat, which is better than that of meat from the domestic animals, and they were not afraid of inadequate health of game meat. Nevertheless, the supply in the market is relatively weak, although the price is acceptable. As many as 85.33% of respondents think that the game meat is poorly marketed and weakly represented in the media. From the above facts, it can be concluded that it is necessary to increase marketing activities and raise citizens' awareness of the nutritional, culinary, and health value of game meat in order to increase the consumption of game meat and improve its position in the market.

Keywords: game meat, market, marketing, consumers, preferences

Stomach content analysis of the Atlantic bluefin tuna in Adriatic Sea

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Summary

Atlantic bluefin tuna, ABT, (*Thunnus thynnus*) is one of the largest marine predators and an important target species for commercial and recreational fisheries. Studies of the feeding ecology are important for better understanding their trophodynamics and the food webs in which they play important role. Stomachs contents of 24 ABT, including both juveniles and adults (weight range from 8 to 80 kg) collected in the eastern Adriatic Sea, were analysed by morphological inventory and DNA metabarcoding. Preliminary DNA metabarcoding tests were performed on five individuals. DNA was extracted from stomach swab and homogenate of stomach contents using a commercially available DNeasy Blood & Tissue kit (Qiagen). Partial fragments of two gene regions (cytochrome c oxidase subunit I (COI) and the ribosomal 18S-V1V2 region) were amplified using universal metazoan primers and commercially sequenced on Illumina MiSeq platform. Molecular identification of prey using DNA metabarcoding approach resulted with greater prey diversity than obtained using morphological approach. A large difference in sequencing reads between the two types of DNA samples, with most reads coming from tuna DNA in the case of the stomach swab was also noted. The most abundant prey species are small pelagic fish (~97%) and cephalopods (~3%). Noteworthy is that we also detected some demersal and gelatinous species such as Atlantic stargazer (*Uranoscopus scaber*), European hake (*Merluccius merluccius*), red mullet (*Mullus barbatus*), smallspotted catshark (*Scyliorhinus canicula*) and *Salpa* spp. These results suggest that ABT is an opportunistic feeder and that DNA metabarcoding can be a powerful tool for analysing the stomach contents of key marine predators.

Keywords: DNA metabarcoding, COI, 18S, morphological analysis, Atlantic bluefin tuna

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Analysis of the current situation and the possibility of improving sheep breeding on the island of Pag

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Summary

Sheep breeding on the island of Pag has a centuries-old tradition and is based on extensive keeping of the autochthonous breed - Pag sheep. The goal of the research was to analyse the current state of sheep farming on the island of Pag. Data from the Croatian Agency for Agriculture and Food (HAPIH) and data from research conducted in the period from 2019 to 2022 were used for the study. Breeding technology, herd health, milk yield and fertility of sheep were analysed. According to HAPIH estimates, the size of the sheep population on Pag ranges from 30,000 to 35,000 heads in 484 flocks. In a survey conducted with 46 randomly selected breeders, it was determined that an average of 48 sheep are milked. The average age of breeders is 59 years. Feeding is based on pasture and supplemented with hay, grains, and concentrates. Although almost 500 breeders of Pag sheep were registered on Pag, production control was carried out on only 35 flocks in 2021. The average length of lactation in controlled herds was 159 days, while the average milk yield was 129 kilograms. In 2022, 199 breeders who deliver milk to cheese factories, were registered which shows that there is no production data for more than 250 farms. According to data from the Central Laboratory for Milk Analysis 654,942 kilograms of milk from Pag sheep was purchased in 2022, and a significantly smaller portion was processed in registered family cheese factories. Based on the number of sheep and average milk yield, it is assumed that only 50% of the total amount of milk is processed into cheese in registered cheese factories, while the rest is sold without supervision. A special problem for cheese makers on Pag is the value added tax on dairy products, as it is one of the highest in the EU. Based on previous research, we can conclude that on the island of Pag there is a possibility of increasing milk yield at herd level with a new approach to breeding and selection work, feeding and reproduction management. Flock health management must be dedicated to eradicating diseases that reduce herd productivity, such as infectious and parasitic diseases.

Keywords: Pag, sheep, production, milk, flock health

The Effect of Season and Dairy Herd Productivity on the Milk Urea Content

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Summary

In dairy cows, milk urea (MU) is an indicator of feeding management with respect to the by-product of dietary protein breakdown. Optimally, cows have 15-30 mg of urea in 100 ml of milk. The aim of our study was to evaluate the effects of season and dairy herd productivity on MU content. The dataset used included 13,524 bulk milk samples from regular AT milk recording collected from 675 dairy farms over a three-year period (2019-2021). Only farms with at least two milk recordings per season in each year (i.e., at least 24 records per farm) and more than three cows per milk recordings were included. The effect of season and productivity on MU was evaluated using a mixed model with fixed effects of season (4-factorial: spring, summer, fall, winter) and herd productivity (5-factorial: < 15, 15-20, 20-25, 25-30, > 30 kg average daily milk yield in the herd) and their interaction, as well as the farm random effect. The MU content increased with herd productivity (from 16.2±8.3 to 21.8±4.9 mg/100 ml), with significant differences between all productivity classes except the two highest. Significant differences were also observed between all seasons; the highest MU content was observed in summer (21.1±7.4 mg/100 ml), followed by autumn (19.6±7.9 mg/100 ml), spring (18.3±6.7 mg/100 ml) and the lowest in winter (17.2±6.5 mg/100 ml). The effect of season was much more pronounced in the low productivity herds. Highly productive herds (average daily milk yield > 25 kg) had more stable MU content during the year (19.5-21.7 mg/100 ml). In contrast, MU content was lower in low productive herds (average daily milk yield < 15 kg) and varied more in different seasons (14.0-19.8 mg/100 ml). The results showed that outside the growing season, many dairy herds, especially low productivity ones, have too low MU contents. In the two lowest productivity classes, almost 50% of the milk samples did not reach the lower limit of 15 mg MU in 100 ml. This is indicative of poor farm management and less controlled/consistent feeding throughout the seasons. To increase MU content, it is recommended to improve the quality of the basic fodder (e.g., sown grasslands with the inclusion of legumes) and to catch the optimal harvesting time.

Keywords: dairy cows, milk urea, productivity, season

Assessment of Risk Factors for Tail Biting in Pig Fatteners: Slovenian Example

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Summary

Tail biting is a serious problem in modern pig production, often occurring because of inadequate housing conditions. To mitigate this behaviour, tail docking is usually used. This invasive method removes the affected body part but does not help to eliminate the causes of tail biting – inappropriate environmental conditions. An alternative approach is to identify the risk factors associated with housing conditions that lead to tail biting and take appropriate action(s). The aim of the study was to determine the extent and severity of tail biting in 16 Slovenian farms (with 30-1000 pigs in rotation) and to assess the risk factors using a detailed questionnaire that provided an accurate overview of the housing conditions. Although all but one breeder raised pigs with docked tails, they occasionally experienced tail biting problems. The survey revealed many risk factors, the most common being inadequate monitoring of (micro)climatic conditions (75%), inadequate pen size (especially at the end of the fattening, 13%) and pen structure (arrangement of pen elements, 83%), lack of litter, use of non-optimal enrichments (74%), inadequate number of feeding places and drinkers (74% and 27%), inadequate feed milling (85%), lack of feed ingredient analyses (54%). Several different risk factors were found to coincide in all the farms studied, but in a unique combination. There are several salient and pervasive risk factors that can be systematically eliminated either through breeder education or legislation adaptation. Since many risk factors could be eliminated with little effort (e.g., lower stocking densities, additional drinkers and feeders, optimal enrichments, feed ingredients control, etc.), significant improvements are already possible if problem solving is approached individually per farm. However, for effective implementation of the basic preventive measures, it is essential that information is consistently transmitted from researchers and other experts to breeders.

Keywords: tail biting, risk factors, assessment, pigs

Teat-cup removal strategies for robotic milking

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Summary

The number of milking robots is rapidly increasing throughout different countries and continents. Mainly they are located in Europe with more than 90% installations. The main difference between milking robot and conventional parlour is the milking interval and t the cows enter the box. It is known that the milking interval increases throughout lactation and therefore take-off setting strategies should be adapted. Automatic cluster removal strategies from the milking parlour cannot be applied in milking robots. Moreover, milking robots differ in pre-milking teat preparation and take-off settings in cows in the stable and on pasture. Many of these differences are associated with cow traffic, pulsator and vacuum settings, feeding, type of box, etc. Appropriate settings of the automatic cluster remover can significantly improve teat condition and parlour or AMS efficiency while maintaining udder health and quantity of milk harvested. In conclusion, the efficiency of robotic milking can be maximized if appropriate management of the dairy herd is applied that motivate the cows to come to the milking box more often voluntarily. Additionally, proper milking teat preparation should be applied together with up to 480 g/min take-off setting which depends on present udder filling of the cow.

Keywords: teat-cup removal, dairy cow, milking robot, milking, udder health

Comparison of Holstein x Belgian Blue and Holstein x Piedmontese crossbreds in beef production: growth, carcass and meat traits

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Summary

The crossbreeding of Holstein cows with beef bulls is used with the aim of increasing progeny value in beef production and improving income from dairy production. Although beef farms recognize the value of these F1 crossbreds, the growth dynamics, carcass traits, and meat quality of crossbreds in conventional beef fattening farms are insufficiently researched. The objective of this study was to determine the influence of genotype and sex on important beef traits in crossbreds of Holstein cows with Belgian Blue and Piedmontese bulls. The study was conducted on 76 F1 crossbred animals, including 39 bulls and 37 heifers. Male calves of both genotypes had significantly ($p < 0.05$) higher birth weights than female. Crossbreed Holstein with Belgian Blue achieved higher slaughter weights compared to crossbreds with Piedmontese ($p < 0.001$). Bulls also achieved higher slaughter weight compared to heifers. Heifers achieved higher carcass fatness ($p < 0.01$). Genotype had a more significant effect on carcass yield ($p < 0.001$) than sex ($p < 0.05$). The influence of genotype ($p < 0.001$) and sex ($p < 0.01$) on the share of bone, muscle and fat tissue in the rib section was observed. The influence of genotype on fat content in meat was also found ($p < 0.01$). Genotype influenced the content of saturated fatty acids in adipose tissue, while the influence of sex on the content of unsaturated fatty acids in adipose tissue was more pronounced ($p < 0.001$). The determined indicators of growth, carcass and meat characteristics can be used in the adjustment of crossbreeding schemes and breeding technology to achieve better carcass and beef quality.

Keywords: crossbreds, Holstein, beef breeds, carcass, beef quality

The application of microencapsulated rennet and autochthonous dairy cultures in the “Paški sir” cheese production

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Summary

The investigation aimed to determine changes in the chemical composition and physical properties during the ripening process of “Paški sir” cheese. The experimental cheeses were produced in three different treatments: (1) cheeses produced with commercial rennet and dairy cultures, (2) cheeses produced with lyophilized autochthonous rennet and microencapsulated autochthonous dairy cultures, and (3) cheeses produced with innovative microcapsules containing autochthonous rennet and dairy cultures. For this purpose, changes in milk fat, protein, dry matter, salt content, and pH values were determined in five ripening stages (0, 30, 60, 90, and 120 days). A total of 18 cheeses were produced, 6 in each treatment. The results show that the changes in determining parameters are not significant ($P > 0.05$) considering treatments ($N=3$). Milk fat, protein, and dry matter content are significantly different considering ripening stages ($N=5$, $P < 0.01$), with their values increasing towards the end of maturation, which is typical for “Paški sir” cheese. Salt content and pH values are significantly different ($P < 0.01$) between days 0 and 30, while there is no difference in the later stages. Based on the obtained results it can be concluded that this innovative application of microencapsulation can be used in “Paški sir” cheese production. This innovative approach can simplify the production process and preserve traditional cheeses’ biodiversity and distinctive organoleptic properties.

Keywords: Paški sir; cheese; microencapsulation; rennet; dairy cultures.

Urea content in milk as an indicator of ammonia emission from dairy cows

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Summary

The aim of the research was to evaluate dietary protein utilization and to predict nitrogen content based on nutritional parameters and milk urea nitrogen content (MUN). It also aimed, to predict the potential of ammonia emission from dairy cows based on MUN and to test whether it is affected by the stage of lactation. The research was conducted under production conditions in four agricultural holdings of different sizes located in various places in the Province of Vojvodina, representing different animal housing and feeding technologies. Test-day records were collected during the regular milk recording performed monthly in accordance with the alternative milk recording method (AT4 / BT4) on dairy cattle farms in Serbia. At each recording, measuring and sampling of milk were performed during evening or morning milking. The statistical analysis was conducted using ANOVA, and correlation and regression analyses. The results emphasized that the optimal amount of urea in milk is 15-30 mg dL⁻¹, and there was a strong relationship between ammonia emission and MUN which was not different among lactation stages. A precise system of protein nutrition for lactating cows must provide a meal in accordance with the level of milk production, but also avoid the consumption of excessive amounts of protein, which leads to their inefficient use, a negative impact on the energy balance of animals, and increased excretion of nitrogen into the environment, but also unnecessary increases the price of meals.

Keywords: milk urea nitrogen, test day record, dairy cattle, ammonia emission

Encapsulation of *Lactiplantibacillus plantarum* in alginate microspheres

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Summary

Lactobacilli have been used as adjunct cultures in the manufacture of different cheeses to accelerate ripening and/or improve cheese quality. One of the most important tasks to be solved in the process of cheese making is adequate protection and timely delivery of the bacterial cultures. *Lactiplantibacillus plantarum*, this bacterial culture is often used in cheese acidification and has a positive effect on the organoleptic properties of cheese. This research aims to explore the possibility of simultaneous encapsulation of *Lactiplantibacillus plantarum* and calcium ions by the ionic-gelation encapsulation method. The process of simultaneous encapsulation in the biopolymer microcapsules was performed under previously optimized conditions. The concentration of sodium alginate was set to 1.5% (w/v), the calcium chloride concentration was 4%, and the vital fraction of *Lactiplantibacillus plantarum* was $9.0 \pm 0.1 \log \text{CFU/g}$. Microspheres were produced and physicochemically characterized (microsphere size, encapsulation efficiency, loading capacity). Simultaneous encapsulation in biopolymer microspheres did not have a negative effect on the *Lactiplantibacillus plantarum* properties. Also, the release date of the bacterial cultures was fitted to the Korsmeyer-Peppas model and the n exponent indicated that the release mechanism was Fickian. The electrostatic interactions between bacterial cultures, alginate, and calcium ions were confirmed by infrared spectroscopy, dynamic light scattering, and zeta potential measurements. The obtained results showed that bacterial cultures and calcium ions can be successfully simultaneously encapsulated and applied in the process of cheese production.

Keywords: encapsulation, *Lactiplantibacillus plantarum*, microspheres, calcium ions, cheese

Simultaneous encapsulation of dairy rennet and calcium ions in alginate microparticles

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Summary

Dairy rennet is a natural curdling enzyme that is used to make cheese. It ensures that the milk curdles without becoming sour. One of the most important tasks to be solved in the process of cheese making is adequate protection and timely delivery of dairy rennet. This research aims to explore the possibility of simultaneous encapsulation of dairy rennet and calcium ions by the ionic-gelation encapsulation method. This is an important step, as the conventional addition of the “free” enzyme is generally not recommended due to its negative influence on the flavor and texture. The process of rennet encapsulation in the biopolymers microcapsules was performed under previously optimized conditions. The concentration of sodium alginate was set to 1.5% (w/v) and calcium chloride concentration to 4%. The final concentration of rennet was set to 20% (w/v). Microparticles loaded with rennet were produced and physicochemically characterized (microparticle size, encapsulation efficiency, loading capacity). Simultaneous encapsulation in biopolymer microparticles did not have a negative impact on the dairy rennet properties keeping its activity very high. Also, the rennet release data were fitted to the Korsmeyer-Peppas model and the n exponent indicated that the release mechanism was Fickian. The electrostatic interactions between rennet, alginate, and calcium ions were confirmed by infrared spectroscopy. The obtained results showed that dairy rennet and calcium ions can be successfully simultaneously encapsulated and applied in the process of cheese production.

Keywords: encapsulation, dairy rennet, microcapsules, calcium ions, cheese

The effects of industrial hemp as a phytobiotic on the production indicators of broiler chickens

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Summary

Hemp (*Cannabis sativa L.*) is a multipurpose plant that has almost zero THC (tetrahydrocannabinol) content. Seeds, hemp seed cake, and hemp oil can be used as supplements to feed mixtures in the feeding of domestic animals. Hemp seeds are rich in crude proteins and crude fats, and the addition of hemp oil and seed cake as well as hemp leaves can affect production indicators and quality of poultry meat and eggs. The aim of the research was to determine the production indicators of broiler chickens fed with feed mixtures containing different proportions of hemp leaves. The research was conducted on 100 male Ross chickens, divided into 4 experimental groups of 25 chickens each. The chickens were fed with starter feed mixtures from the 1st to the 21st day and with the grower feed mixture from the 22nd to the 42nd day. The feed mixture for the control group (K-0) did not contain hemp leaves, while they were mixed in the feed mixtures for the experimental groups (P-1, P-2 and P-3) with a proportion of 1, 2 and 3%. Control weighing of chicks was carried out on the first day and at the end of the 1st, 2nd, 3rd, 4th, 5th and 6th weeks of the age. In the same intervals, the amount of feed mixture consumed was determined and conversion was calculated. At the age of three weeks, the average body weight of groups K-0, P-1, P-2 and P-3 was 1037.00; 1021.92; 933.50 and 973.16 grams, respectively, and the weights of chickens in groups K-0 and P-1 were statistically significantly different ($p < 0.05$) from those in groups P-2 and P-3. The conversion of starter feed mixture was 1.22; 1.29; 1.26, and 1.24 kg/kg. At the end of the research, the negative impact of the addition of hemp to the feed mixtures of broiler chickens on body weights continued, which was 2979.12; 2626.57; 2429.16; 2630.88 grams and all weights of the experimental groups (P-1, P-2 and P-3) were statistically significantly lower ($p < 0.05$) compared to the control group (K-0). Also, the achieved conversion of the feed mixture of the control group (K-0) 1.54 kg/kg during the entire research was lower than the conversion of the feed mixture of the experimental groups (P-1, P-2 and P-3) 1.73; 1.76 and 1.65 kg/kg. The obtained results indicate that hemp leaves (*Cannabis sativa L.*) in the proportion of 1, 2 and 3% in feed mixtures cannot be recommended for feeding broiler chickens because it has a negative effect on the production results of fattening.

Keywords: hemp (*Cannabis sativa L.*), production indicators, broilers

Meat production indicators of roosters of Posavska crested hen

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Summary

The goal of this research was to determine production indicators in meat production, slaughterhouse indicators and morphometric properties of posavska crested hen. The research was conducted on 5 strains of posavska crested hen breed: yellow vetch, vetch, red colored, lavender and golden brown. Feeding was *ad libitum* for the whole time, adapted to the age of chickens from nutritional point of view. Separation by gender was performed at the age of 4 weeks and the slaughter of roosters and determination of slaughterhouse indicators at the age of 20 weeks. The average body weight of roosters was 3319.97 g and 2420.2 g of pullets at the age of 18 weeks. The largest mass was achieved by roosters of lavender strain (3542.63 g) and lavender pullets (2562 g). The average conversion of the feed mixture at the age of 4 weeks was 2.79 and at 18 weeks it was 3.8 for roosters and 4.79 for pullets. The lowest conversion at 4 weeks of age was achieved by lavender strain (2.39), at 18 weeks of age the lowest conversion was with the roosters of the lavender strain (3.77) and pullets of the golden brown strain (3.81). Average mortality was 16.25% for roosters and 11.6 for pullets. There was no mortality for yellow vetch, vetch and golden brown strain of roosters. The average carcass yield was 71.54% and the highest was with the red colored strain 72.51%. The proportion of breast with bones was the highest with vetch and red colored strain (23.55%), the proportion of fillets was the highest with lavender strain (15.64%), the proportion of drumsticks was the highest with vetch strain (17.64%) and the proportion of gables was the highest with the yellow vetch strain (18.56%). With flesh color the highest value of light intensity L* had yellow vetch strain (70.32) and value a* (5.70) and b*(5.11) was with the lavender strain. Morphometric properties did not differ significantly between strains.

Keywords: Posavska crested hen, production indicators, slaughterhouse indicators, morphometric properties

Variability of intramuscular fat and fatty acids in Black Slavonian Pig

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Summary

The Black Slavonian pig (BS) is the most numerous autochthonous pig breed in Croatia, whose number has been continuously growing in the last 20 years. Many studies have shown that the meat of BS is of exceptional quality compared to the meat of modern breeds. The aim of this research was to investigate the influence of acorn feeding on intramuscular fat (IMF) and fatty acids profile. This research included a total of 80 BS fatteners equally aged, fed different diets until their final body weight. Therefore, the fatteners were divided into two groups of 40 animals. One group was fed with classic feeding mixtures until the end of the fattening period, while the other group had 3 kg of acorns per animal added into the diet (28 days before slaughter). The determination of IMF was analyzed by Soxhlet extraction method and the NIR method. Fatty acid profiles were determined by gas-liquid chromatography according to the reference ISO method. The day before slaughter, the animals weighed an average of 141.50 kg, while the average dressing percentage was 83.5%. Higher average IMF value was determined in pigs fed with classic feeding mixtures (5.02%), while in pigs additionally fed with acorns, the IMF value was average 3.93%. Statistically significant difference ($P < 0.05$) between the groups was found for the content of unsaturated fatty acids (UNSFA).

Keywords: Black Slavonian pig, carcass weight, IMF, SFA, UNSFA

Drones in animal husbandry

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Summary

With the development of unmanned aerial vehicles (UAV, drones), new technologies are available to farmers, especially those involved in animal husbandry, to assist them in production. The aim of this thesis is to go over the most recent technological advancement in drone hardware and software that have practical applications in animal husbandry. One of the biggest problems faced by farmers, especially when they have a large herd of cattle (e.g., more than a thousand cows for example), is that a person has great difficulty tracking each individual animal. Cattle herds that are grazing on large pastured field can be difficult to manage. Health and safety of each animal, as well as the quality of the grazing pasture could be easily monitored through drones with cameras and other sensors. With a bird's eye view of larger pastures, one person could monitor a larger herd over greater distances. Special sensors like thermal vision can help determine animals' health and assist in search and rescue operations in low light conditions. Multispectral and hyperspectral cameras that are able to "see" the invisible spectrum of light can be used to determine crop and pasture health. Deep learning machines can use pictures to "learn" animal behavior and migration patterns as well as detection of different animal species that are moving through the farm and pasture. Speakers, microphones, and searchlights can be attached to the frame of a drone, and with this setup, farmers can steer their herds in the desired direction. There are a number of ways drones and sensors can be used in livestock production. This thesis went through the most common and versatile practical usages of what that farmers can profit from today

Keywords: drone, multispectral, hyperspectral, deep learning machine

Genomic patterns of homozygosity in Istrian sheep

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Summary

The runs of homozygosity (ROH) pattern can provide useful information about the history and genetic structure of livestock populations. The aim of this study was to determine the ROH profile and level of inbreeding in the Istrian sheep using ROH (F_{ROH}) and information on observed and expected homozygosity (F_{HOM}). The subpopulation under study comprised 719 animals genotyped with the OvineSNP50 BeadChip. Analyses were performed using the PLINK toolset (v1.09). After quality control (MAF<0.05; call rate per SNP 0.10 and animal 0.05), 656 animals and 38,351 SNPs were included in the analyses. The minimal number of SNPs in ROH was set to 44, the maximal gap between SNPs was set to 1 Mb, the minimum SNP density was set to 1 SNP every 250 kb, no heterozygotes allowed, no missing SNPs allowed less than 4 Mb, and the minimum length that constituted the ROH was set to 1 Mb. A total of 15,817 autosomal ROHs ranging from 1.9 to 70.1 Mb were detected. On average, there were ~24 ROHs per animal with an average length of ~8.12 Mb. The frequencies of ROH_{1-4 Mb}, ROH_{4-8 Mb}, ROH_{8-16 Mb}, ROH_{>16 Mb} were 5.6%, 51.4%, 31.4% and 11.6%, respectively. The total length of ROH_{1-4 Mb}, ROH_{4-8 Mb}, ROH_{8-16 Mb}, ROH_{>16 Mb} in the total length of the genome covered by ROHs was 9.4%, 29.2%, 34.5% and 26.9%, respectively. As suggested for medium-density SNP data, only large ROH segments (>4 Mb) were used to assess inbreeding ($F_{ROH>4Mb}$). The average genomic $F_{ROH>4Mb}$ and F_{HOM} were 0.075 and 0.060, respectively, and the correlation between them was high ($r=0.976$). The estimated level of genomic inbreeding in this population calls for more attention in planning future matings to decrease inbreeding below the conventionally accepted level (6.25%). The results provide a reliable source of information for developing basic guidelines for implementation of optimum contribution selection.

Keywords: Istrian sheep, runs of homozygosity, genomic inbreeding coefficient

This research was funded by the Croatian Science Foundation (Genomic characterization, preservation, and optimum contribution selection of Croatian dairy sheep, OPTI-SHEEP), under grant number IP-2019-04-3559.

Influence of selenium addition from different sources on immunoglobulin concentration in plasma of weaned piglets

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Summary

The goal of the research was to determine influence of Selenium (Se) addition from different sources on immunoglobulin G (IgG) plasma concentration in weaned piglets. Piglets (PIC) were weaned on day 30, randomly allocated 10 per group (equal numbers of females and males) and fed with balanced feed for weaned piglets for 42 days. The control group (C) was fed with no Se added in the premix, the group (BiOSe) was fed with Se biofortified cereals (corn and wheat) and the group (CHE) was fed with a chelated version of Se (Framelco®, Switzerland) in the premix. The Se content in the feed was balanced to 0.3 ppm in the experimental groups. Blood was collected and plasma separated on day 1, 21 and 42 of the experiment. IgG concentration in plasma was determined by clinical chemistry analyzer AU 400 (Beckman Coulter, Germany) using Beckman Coulter reagents. Normality of distribution was checked by the Kolmogorov-Smirnov test. Repeated measures ANOVA GLM procedure was used to test the influence of day, group, sex, and interactions. Related to sex, higher ($P < 0.05$) concentration of IgG was determined in plasma of male (3.35 g/L) compared to female piglets (2.67 g/L). This points to either better humoral defense or higher exposition to antigens in male weaned piglets. Similar goes for piglets' age: higher ($P < 0.05$) concentration of IgG in plasma was determined on 42nd day (3.89 g/L) of experiment compared to 21st (2.71 g/L) and 1st day (2.48 g/L). The interaction Day x Group showed higher plasma IgG concentration in BiOSe group (4.77 g/L) compared to C group (3.07 g/L) on day 42 of the experiment. This could mean that piglets fed with Se biofortified cereals have better humoral defense compared to the control group. Addition of biofortified cereals to the diet of weaned piglets increases humoral immune response on day 42 of the experiment, which was not visible on day 21.

Keywords: immunoglobulin, weaned piglets, selenium, biofortification

Mitochondrial DNA analysis in livestock breeding

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Summary

Mitochondrial DNA (mtDNA) sequence is maternally inherited and is one of the most commonly used genetic markers. Since mtDNA has a high level of variability, it is widely used in genetic variability studies and can often be traced back to pre-domestic populations. In addition to tracing the maternal origin of populations back to ancient times, mtDNA variation studies can be particularly useful for establishing relationship between domesticated species and their wild relatives, for identifying sites of domestication, and also for studies of genetic variability within and between populations. Determination of mtDNA haplotypes within the population and comparison with haplotypes of other phenotypically closer and more distant breeds can be used as one of the methods for genetic assessment of the population in the process of autochthonous breed recognition which we currently use for genetic determination of the Dalmatian Bušak horses. The aim of determining mtDNA haplotypes in the population of Banija spotted pigs is to define and preserve maternal lineages which will contribute to the conservation of genetic variability and sustainable breeding of this small population of autochthonous pigs. In populations where herd books have been maintained for centuries and where maternal lineages are very well defined through pedigree data, such as in Lipizzan horses, mtDNA sequencing can be used to check for more or less accidental mistakes in pedigree data.

With the purchase of a genetic analyzer, we have enabled the service of mitochondrial DNA sequencing in the genetic laboratory of the Croatian Agency for Agriculture and Food, and now we can meet the wishes and needs of our breeders in this segment as well.

Keywords: mitochondrial DNA, autochthonous breeds, genetic variability, verification of pedigree data

Comparative estimation of inbreeding on the X chromosome in three native Croatian cattle breeds

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Summary

Estimation of inbreeding is important for the survival of genetically small populations. Therefore, the objective of this study was to i) estimate genomic inbreeding coefficients on the X chromosome in three native Croatian cattle breeds, ii) compare estimates of different genomic inbreeding coefficients on the X chromosome based on identity by descent (IBD) with those based on identity by state (IBS), and iii) analyse segmental variation in inbreeding on the X chromosome. The analysis was based on three Croatian cattle breeds (112 individuals) genotyped with the Illumina BovineHD SNP chip (777,000 SNPs). Our analyses revealed peculiarities in the estimation of genomic X chromosome inbreeding compared to autosomal inbreeding, e.g. higher variation and higher level of inbreeding, which should not be neglected. The obtained results can be used to estimate inbreeding depression and to identify deleterious mutations related to the X chromosome. However, to obtain more general knowledge about the genomic inbreeding level of the X chromosome, more cattle breeds should be analysed.

Keywords: inbreeding, X chromosome, cattle, high density SNP data, small populations

Acknowledgements: This study was supported by project ANAGRAMS-IP-2018-01-8708 funded by the Croatian Science Foundation and by the project KK.01.1.1.04.0058 funded by the EU Operational Programme Competitiveness and Cohesion in the period 2014–2020.

Application of optimal contribution selection in Landrace and Large White pigs in Croatia

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Summary

Selection for economically important traits in pigs is based on methods that require pedigree information. Such an approach results with the choice of related animals for parents of future generations, resulting with increased inbreeding rate in the population and impaired genetic diversity. Using optimal contribution selection balance between genetic improvement and preservation of genetic diversity. The aim of the study was to apply optimal contribution selection algorithms on Landrace and Large White pigs in Croatian national breeding programme. The data set contained around 134K litter records for Large White, Landrace, and their crosses and around 52K animals in pedigree. Optimisation algorithms were applied in two scenarios: the first scenario was based on the maximisation of genetic gain and restricting the mean kinship in the offspring accounting also for breeding values. The second scenario was based on minimising inbreeding by restricting the average kinship. In the first scenario, different kinship constraints between candidates resulted in changing the number of selected animals and changed average breeding values, so that the number of selected candidates increased when the kinship constraint was stronger, with a simultaneous decrease in breeding values. The second scenario resulted in increased inbreeding when the additional weight on genetic gain was added. The use of optimal contribution selection in the pig breeding programme in Croatia is possible with additional efforts to improve pedigree and data quality.

Keywords: pigs, selection, optimal contribution

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Overview of mycotoxin occurrence in cereals: a ten-year Croatian survey

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Summary

Mycotoxins are secondary fungal metabolites expressing carcinogenicity, teratogenicity, immune toxicity, neurotoxicity, hepatotoxicity, nephrotoxicity, reproductive and developmental toxicity in numerous species. They can be found in a variety of important agricultural products dependent on moisture content, water activity, relative air humidity, temperature, matrix composition, and the degree of physical damage. Research on mycotoxin occurrence in cereals as the main contamination source, is essential for developing preventative and management strategies that aim to protect human and animal health. In this study, the occurrence of major storage (aflatoxin B₁ (AFB₁) and ochratoxin A (OTA)) and field mycotoxins (*Fusarium* mycotoxins deoxynivalenol (DON), zearalenone (ZEN), fumonisins (FUM), and T-2/HT-2 toxins (T2/HT2)) in most important Croatian crops, i.e., maize, wheat, barley, and oat (4,080 samples) was investigated over 10 years (2010 - 2020). Contamination with carcinogenic AFB₁, most concerning from the public health standpoint, was determined in 10% of samples during 2013, and is to be attributed to the interchange of extremely dry and extremely hot periods witnessed in 2012 during maize cultivation and to the inappropriate storage. *Fusarium* mycotoxins were frequently uncovered in different cereals throughout the study period, the positives spanning from 42% (for T2/HT2) to even 85% (for FUM). After heavy rains, such as in 2015, the occurrence had risen significantly ($p < 0.05$). Bottomline, Croatian cereals are mainly contaminated with *Fusarium* mycotoxins, but weather extremes had been proven to enhance AFB₁ contamination, too. Therefore, climate changes can not only aggravate mycotoxin contamination documented insofar, but also introduce novel mycotoxins into the region. Further research should identify mycotoxigenic mould growth enhancers and efficient measures to reduce contamination of cereal-containing foods and feeds.

Keywords: mycotoxins, cereals, contamination, Croatian fields, aflatoxin B₁, *Fusarium* mycotoxins

Functional and health properties of 10-HDA acid – a unique component of the royal jelly

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Summary

Royal jelly is a special biologically active substance, with an extremely complex chemical composition, and is produced as a product of worker bees. The basic ingredient of royal jelly is 10-hydroxy-2-decenoic acid (10-HDA). It belongs to the group of unsaturated fatty acids and is not found anywhere in nature and cannot be synthesized. This is precisely why it is used as an indicator of the authenticity and quality of royal jelly products. The tradition of using royal jelly is centuries old and various pharmacological effects are attributed to it, such as antitumor and antibiotic activity and strengthening of the immune system. For these reasons, royal jelly is widely distributed commercially and is used for medicinal purposes all over the world as fresh and/or in freeze-dried form. The aim of the work is to analyze products with royal jelly on 10-HDA, to compare with the declared values on the label and health claims, and to relate them to the functional and health properties that stand out. The analysis is carried out on HPLC and results vary from 1.5% to 3.5%.

Keywords: royal jelly, 10-HDA, functional properties, health impact, legislative framework, market of the Republic of Croatia

EU SCHOOL SCHEME: Attitudes of school children and their parents about the importance of milk in the diet

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Summary

Upon joining the EU in 2013, the Republic of Croatia joined many EU countries implementing the EU Joint Action School Fruit and Vegetable Scheme, and a few years later began implementing the Milk Program - free meals with milk and dairy products for primary school children. The main goal is to educate students about the importance of a healthy diet and distribute free fruit and vegetables, milk and dairy products. Each year, surveys are conducted among school children and their parents about the project. The results of the surveys conducted over the years show that only 50% of the students drink a cup of milk every day. 40% of students drink milk and dairy products most often in the morning and 19% in the evening. Plain milk and chocolate milk are preferred by 26% of students, and surprisingly, fruit yogurt is preferred by only 13% of students. Through education and offering free meals with milk and milk products, students create a positive attitude and increase their consumption, and considering the accelerated growth, milk consumption has a positive impact on students due to its nutritional composition.

Keywords: School scheme, dairy products, students, parents, primary school

Development of meat products with reduced amount of nitrites and nitrates

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Summary

In the European Union all food additives are identified by E numbers. They are safe, well-studied and evaluated for harmful side effects. However, even today, there is substantial consumer distrust towards additives and their labeling. Because of this a need arises to replace additives like potassium or sodium nitrates and nitrites, which are the ones most commonly used in meat products, with natural sources. The aim of this research is development of meat products without added nitrites and nitrates. Different kind of meat products are studied: bologna sausage, cooked ham, mortadella and fermented sausage. In thermally processed products the focus is on replacing added nitrite salt with yeast extract and bacterial culture. In the case of fermented sausage, added nitrite salt and sodium nitrate are replaced with sea salt and bacterial culture. The other part of the project is focused on addition of natural source of nitrates that are found in vegetables, together with bacterial culture. The statistical analysis of the results for ham shows a P-value <0.001 between recipes with added nitrite salt and alternative recipes. The value of the median mass concentration of nitrite in the recipe with added nitrite salt is 23.11 mg/L, whereas this value is below the limit of quantification in alternative recipes. Also, for other meat products, the P-value confirms the hypothesis for replacement of nitrites with natural sources.

Keywords: additives, nitrates, nitrites, meat products, natural sources of nitrates

Some properties of kefir obtained from different ratios of pineapple juice-milk mixtures

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Summary

Kefir is a product that has important effects on health, but not preferred by many consumers. In this study, it was aimed to develop a product especially for individuals who do not consume kefir. For this purpose, 5 different kefir samples were produced from mixtures of pineapple juice (K-0%, A-5%, B-10%, C-15%, D-20%) and cow's milk, and their properties were investigated. While there was no difference between the dry matter contents of the samples, the protein, fat and ash contents decreased with the fruit juice addition. The consistency index values decreased with the increase in the amount of fruit juice; furthermore, there was no difference in flow behavior index values and all samples showed non-Newtonian behavior ($R^2 > 0.99$). In addition, there was a slight increase in the phase separation values with the addition of fruit juice. The lactic acid bacteria, total mesophilic aerobic bacteria and yeast-mold counts increased in the samples with the increase in the amount of fruit juice, possibly related to the high content of carbohydrates. Sensory evaluation of the samples (appearance, texture, flavor, total acceptability) was carried out by two different experienced groups of 7 panelists each. The control sample got the highest scores in terms of all properties by the group with the habit of consuming kefir. On the other hand, the panelists, who do not ordinarily consume kefir, gave higher scores to the samples with fruit juice; and the samples containing 10% and 15% fruit juice got the highest scores in terms of flavor and total acceptability. It was concluded that the kefir samples (especially those containing 10% and 15% pineapple juice) examined in the study may be an alternative product for consumers who do not have the habit of consuming kefir.

Keywords: kefir, pineapple juice, rheology, lactic acid bacteria, sensory evaluation

Modelling of tailor-made solvents for sequential extraction of bioactive compounds from myrtle leaves

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Summary

Myrtle (*Myrtus communis* L.) is an aromatic medicinal plant rich in bioactive compounds, which gives it great potential for industrial use. While the efficiency of extraction, being an essential step in the utilization of these compounds, can depend on many factors, the choice of the extraction solvent is critical. Since solubility is based on the similar polarity of solvent and solute, different groups of compounds are extracted with solvents of the appropriate polarity. Sequential extraction can therefore be used to extract different chemical groups from the same plant material. It is based on the separation of the plant material depleted with one solvent and its subsequent extraction with a solvent of a different polarity. Therefore, in this study, aqueous solutions containing 0-96% ethanol were used for the extraction of pigments and phenolic compounds from myrtle leaves. The behavior of total chlorophylls was best predicted by an exponential model, while carotenoids were characterized by a sigmoidal curve. Both chlorophylls and carotenoids reached their highest yield in aqueous solutions containing 96% ethanol. The phenolic compounds were fitted through a second-order polynomial model, with the highest yield obtained with 30% ethanol in water. Overall, higher yields of pigments and phenols are obtained in sequential extraction when a 96% ethanolic solution is used as the first solvent and a 30% ethanolic solution is used as the second solvent.

Keywords: sequential extraction, myrtle, chlorophylls, carotenoids, phenols

Strawberry tree fruit (*Arbutus unedo* L.) and 3D printing – Development of an innovative functional food product

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Summary

Strawberry tree fruit (*Arbutus unedo* L.) is an excellent source material for the production of functional foods. This is because of the valuable bioactive compounds abundant in this fruit, which are believed to have many health benefits. Therefore, the aim of this study was to investigate the possibility of using strawberry tree fruit in the production of 3D printed snacks through additive manufacturing. The influence of two different 3D printing (3DP) programs as well as the type (corn vs. wheat) and the level of added starch carrier (4, 6 and 8%) were analyzed. All samples were analyzed for water activity (a_w), pH, mass fraction of bioactive compounds, pigments, and antioxidant capacity. The type of starch carrier had a statistically significant effect on a_w , pH, total phenolic compounds, hydroxycinnamic acids, flavonols, monomeric anthocyanins, condensed tannins, total carotenoids, and antioxidant activity of the 3D-printed samples. The lower level of starch carrier (4 vs. 6 and 8%) had a positive effect on the stability of the analyzed bioactive compounds, pigments and antioxidant capacity. The type of 3DP program had a statistically significant effect on the mass fraction of flavonols, chlorophyll a and b, total carotenoids and on the antioxidant activity of the 3D printed samples. In conclusion, 3DP technology can be considered as promising in the field of development of innovative and functional strawberry tree fruit-based products.

Keywords: functional food, strawberry tree fruit, 3D printing, starch type, bioactive compounds

The use of grape pomace in food products - an overview

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Summary

The main byproduct in wine production is grape pomace, which consists of stems, skin, pulp and seeds remaining after grape processing. The wine sector, like others, needs to reduce its impact on the environment, and one of the ways to achieve that is a greater use of by-products and waste reduction. Grape pomace, as a valuable by-product in wine making, is characterized by high water content (55-75 %), which depends on the processing conditions. Pomace primarily consists of 43-75 % dietary fiber, 6-15 % protein, lipids and a wide variety of polyphenolic compounds, the most common of which are anthocyanins and flavanols. Abundance in bioactive compounds and the ever-growing requests of the consumers for novel, functional food, have led researchers to find new uses for grape pomace. Up till now, several food products have been successfully enriched with polyphenols from grape pomace: biscuits, flour, dairy products and others. This paper presents an overview of the food products where grape pomace has successfully been utilized - from one side in the form of flour (bakery industry) and from the other side as extracts (dairy industry, drinks etc), as well as some new ideas where pomace can be used "as is" in raw food products.

Keywords: grape pomace, food, polyphenols, food waste

Acknowledgement: This research was financed by the European Regional Development Fund, Operational Programme Competitiveness and Cohesion 2014.-2020. (project KK.01.1.1.07.0007 „Sustainable waste management of winery by-products“)

Viral contaminants of bivalve molluscan shellfish harvested from production areas in Croatia: the results of a three-year study

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Summary

Bivalve molluscan shellfish (BMS) are prone to microbial contamination due to their filter-feeding nature. Whilst bacterial contamination is being monitored by *E. coli* count, viral contamination is overlooked. Nevertheless, the outbreaks in the community are occurring and their frequency is probably underestimated. Our study aimed to identify the extent of BMS contamination by *norovirus* (NoV), *rotavirus A* (RVA), *hepatitis E virus* (HEV) and SARS-CoV-2 in Croatia. During three years (2018-2021) 275 digestive tissue samples (2 g pools of hepatopancreas) were collected from three species of BMS (Mediterranean mussels, N= 203; European flat oysters, N= 61; and warty venus clams, N= 11) harvested from commercial production areas (N= 22) on the Croatian side of the Adriatic Sea. The virus concentration procedure and RNA extraction were conducted according to the standard ISO 15216-1 2017, using Mengovirus as a whole process control. The NoV (GI and GII), RVA, HEV, SARS-CoV-2 (on 77 samples collected in 2020/2021) and Mengovirus were detected by RT-qPCR. The results reveal the anthropogenic influence with NoV detected in 30.8% (GI 10.9% and GII 26.3%) and RVA in 23.3% of BMS samples. Mussels were the most affected by viral contamination (NoV 37.2% and RVA 30.6%), followed by oysters (NoV 15.3% and RVA 1.7%) and clams (NoV negative and RVA 9.1%). Nevertheless, no evidence of HEV and SARS-CoV-2 circulation in BMS was observed. The genomic quantities of RVA and NoV were mostly up to 300 gc/g of digestive tissue. Seasonal variations were observed, especially regarding NoV with the highest contamination rate (60% NoV positive BMS samples) observed in the winter 2018/2019 which corresponded to the outbreak reported in humans at that time. The outbreak of NoV GII was connected to the consumption of contaminated BMS from two production sites in Southern Croatia. Our study emphasizes the importance of regular monitoring of BMS for potential viral contamination, especially concerning NoV and RVA. On the contrary, low public health risk was observed regarding SARS-CoV-2 and HEV.

Keywords: bivalve molluscan shellfish, Croatia, norovirus, rotavirus A, SARS-CoV-2, HEV

Maize grain hardness influences faecal parameters and dry matter *in vivo* digestibility in pigeons

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Summary

Maize (*Zea mays* L.) grain is the main feed ingredient in domestic pigeon (*Columba livia* var. *domestica*) nutrition. However, data on the digestibility of maize grain in pigeons are scarce and insufficient for modern pigeon nutrition. The aim of this study was to investigate the *in vivo* dry matter digestibility (DMD), faecal pH, organic acid content and microbiota in pigeons fed three maize hybrids differing in grain hardness. The tested hybrids differed in grain hardness (flotation index from 53 to 97%, breakage susceptibility from 70.16 to 88.94%, Stenvert coarse-to-fine ratio in milled grain from 0.308 to 1.005), and the hardest hybrid had higher DMD compared to the softest hybrid (88.40% vs. 84.69%, $P < 0.05$), but not compared to the hybrid with medium hardness and medium DMD (87.55%, $P > 0.05$). In addition, feeding with the hardest hybrid had more favourable faecal characteristics ($P < 0.05$): lower but not too low pH (7.11) and higher lactobacilli count ($10.10 \log_{10}$ cfu/g) compared to the other two hybrids. (pH: 7.77 and 7.30; lactobacilli count: 8.59 and $8.96 \log_{10}$ cfu/g). Salmonella was not detected in any faecal sample. Acetic acid content tended to increase with maize hardness ($P = 0.07$), with a content of 1.74 g/kg DM in the hardest hybrid compared to 1.22 and 1.49 g/kg DM in the other two hybrids. This study shows the importance of physical characteristics in the selection of suitable maize hybrids for pigeon nutrition, and that harder hybrids result in higher digestibility and more favourable faecal characteristics.

Keywords: maize, grain hardness, pigeons, dry matter digestibility, fecal microbiota, fecal pH and organic acids

Production characteristics and quality of eggs from laying hens fed diets differing in commercial maize hybrid

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Summary

The proportion of maize hybrids in the complete feed can be up to 70%, so even small differences in physicochemical properties between hybrids can affect the production characteristics of laying hens. The aim of this study was to investigate the effects of high-yielding maize hybrids on hen production traits and egg quality. Treatment diets contained 60% of grain and differed in only 15 commercial maize hybrids from six seed companies. By three in each cage, 225 Lohmann Brown hens were allocated to 15 dietary treatments in a completely randomised design (15 treatments×5 cages). The experiment lasted 12 weeks, during which egg number and weight were recorded daily, and diet intake was recorded weekly. Eggs for quality analysis were collected once a week during the last five weeks of the experiment. In terms of production characteristics, diet intake and egg production were similar among treatments, while treatments differed ($P<0.05$) in average egg weight (58.04-61.11 g), average daily egg mass (52.55-60.68 g) and feed conversion ratio (2.01-2.45). For egg quality parameters, egg albumen height, Haugh units and shell thickness were similar between treatments, while treatments differed ($P<0.05$) for yolk colour fan (6.28-8.76) and shell strength (4.54-5.20 kg/cm²). The obtained results suggest that differences between hybrids could lead to different feed conversions and egg properties which could affect farmers' overall income.

Keywords: maize hybrid, laying hen, production characteristics, egg quality

Acknowledgments: This work has been fully supported by Croatian Science Foundation under the project ColourMaize (IP-2019-04-9063). The work of doctoral student Dora Zurak has been fully supported by the "Young researchers' career development project – training of doctoral students" of the Croatian Science Foundation.

Carotenoid deposition efficiency in egg yolks of laying hens fed diets differing in maize hybrid

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Summary

Yellow maize is the most used component of the hen diet with significant carotenoid content, and its ability to pigment the egg yolk varies among hybrids. Therefore, this study aimed to evaluate the efficiency of carotenoid deposition in the yolk of hens fed diets differing in 15 maize hybrids from six seed companies. By three in each cage, 225 Lohmann Brown hens were allocated to 15 dietary treatments in a completely randomised design (15 treatments×5 cages). The experiment lasted 12 weeks, and eggs for analysis were collected once a week for five weeks after stabilising total carotenoid (TC) content. The carotenoid profile of diets and yolks was determined using HPLC method, and the deposition efficiency of individual and total carotenoids was calculated as the ratio between carotenoid production in the yolk and carotenoid consumption by the diet. Dietary treatments varied in carotenoid profile in both diets and yolks ($P < 0.001$), resulting in TC content from 10.50 to 24.31 $\mu\text{g/g DM}$ and 20.40 to 43.03 $\mu\text{g/g}$, respectively. The deposition efficiency of lutein and zeaxanthin was higher than that of α - and β -cryptoxanthin and β -carotene in all dietary treatments (average 24.97 vs. 9.17%). Dietary treatments differed in the deposition efficiency of lutein, zeaxanthin, β -carotene, and TC ($P < 0.001$), which ranged from 18.72 to 25.58%. The obtained results suggest that some hybrids are more efficient in depositing carotenoids into the yolk and therefore are more suitable for hen nutrition.

Keywords: maize hybrid, carotenoids, deposition efficiency, laying hen, egg yolk

Acknowledgments: This work has been fully supported by Croatian Science Foundation under the project ColourMaize (IP-2019-04-9063). The work of doctoral student Dora Zurak has been fully supported by the “Young researchers’ career development project – training of doctoral students” of the Croatian Science Foundation.

Tradition versus reformulation: A case study on Czechoslovak sausage recipe „Gothajský salám“

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Summary

The aim of the study was to describe quality parameters and innovate the traditional recipe of the well-known cooked salami “Gothajský salám” in terms of nutritional value and attractiveness for today’s consumers. The quality requirements for the traditional sausage are defined in a national standard (CSN 577231), and currently the quality requirements are specified in Decree No. 69/2016. According to this, “Gothajský salám” is made from pork and beef, with a meat content of more than 40% and a fat content of no more than 40%. It has an elastic and cohesive consistency, a pink colour and irregularly arranged fat grains up to 8 mm in diameter. The study summarizes the results of four diploma theses of four students from Mendel University in Brno (2018-2022). Several groups of products with different grain size (> 8 mm, < 8 mm), salt content (1.6%, 1.8%, 2.0%) and type of seasoning blend (without and with glutamate) were prepared. The acceptability of the novel products to different consumer groups was observed. Two groups of subjects were used for the sensory analyses. One group consisted of elderly participants older than 60 years (n = 119) and the other consisted of students from the Faculty of Agrisciences younger than 30 years (n = 120). It was found that “Gothajský salám” according to traditional recipe was better evaluated by the older generation of consumers (P < 0.05). The younger consumers focus more on other meat products, especially lean ham and frankfurters. Thus, the younger participants were not aware of the existence of the standard for production compared to the older participants (P < 0.05). The mosaic of the product formed by the bacon was perceived as less healthy, and when a finer grain was used the products were acceptable to the younger generation. It was found that the salt content can be significantly reduced to 1.6%, but only when seasoning blend with glutamate is used (P < 0.05). It can be concluded that the traditional “Gothajský salám” recipe could be innovated to meet the demands of a younger generation of consumers if a finer grain is used in the preparation. The salt content can be reduced to 1.6%, but high-quality ingredients and glutamate should be added.

Keywords: cooked salami, food quality, sensory analysis, salt reduction, monosodium glutamate

Effect of the addition of brown wakame seaweed (*Undaria pinnatifida*) on the quality of dried cooked and dried fermented sausages

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Summary

The reformulation of recipes for meat products is a hot topic in many European countries. On the one hand, the protection of traditional food recipes is being resolved and regional production is being promoted. On the other hand, the impact of processed meat and its consumption on human health is being clarified. In general, meat products are considered to have a negative impact on human health. The main problems are the fat and saturated fat content, the high salt content (sodium) and finally the use of additives. To some extent, these are incompatible attitudes within the food quality and nutrition strategy for the future. Other issues include sustainability, efforts to reduce the impact of the meat industry on greenhouse gas emissions, reduction of byproducts, or the use of natural substances with technological or nutritional value found in natural materials. Seaweed could be a good source of nutrients and an element to improve the formulations of meat products. By adding seaweed to meat products, new, healthier meat products can be produced. The aim of this study was to compare the quality parameters of the reformulated meat product Spišská klobása (cooked and dried sausage) and Dunajská klobása (fermented and dried sausage) with the addition of 1% and 3% wakame algae on chemical, microbiological and sensory quality characteristics. Compared to the control group, the percentage of fatty acids in the experimental meat products did not change significantly ($P > 0.05$). The color results indicate a visible color change to dark, with lower L^* ($P < 0.05$). Microbiological analysis showed that the reduction in salt content had no effect on the microbiological quality of the sausages ($P > 0.05$). It is concluded that the addition of wakame seaweed had no negative effect on the quality characteristics of the sausages ($P > 0.05$), despite the change in color. However, the properties essential for food quality were maintained and the nutritional value did not change.

Keywords: algae, meat product, colour, food safety, sensory analysis, food microbiology

The chemical properties of corn extrudates enriched with agrofertilized wheat

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Summary

The aim of this paper was to determine the impact of the extrusion process on chemical composition, total starch content, starch damage and the bioaccessibility of zinc (Zn) and selenium (Se), using different extrusion temperature profiles: 140/170/170 °C, 150/180/180 °C, 160/190/190 °C and adding different content of agrofertilized wheat with Zn and Se to corn grits: 10, 20, 30 and 40%. The protein and mineral content of the wheat fortified with Zn and Se was significantly higher compared to the corn grits, and the wheat fortified with Zn had the highest protein content ($14.28 \pm 0.76\%$) of all observed raw materials. Starch content increased after the extrusion process with no significant differences with increasing extrusion temperature, while the addition of different proportions of wheat fortified with Zn and Se decreased starch content. In addition, the extrusion process significantly increased the starch damage without significant change by increasing extrusion temperature. Addition of different proportions of wheat fortified with Zn and Se reduced starch damage values in proportion to the added content of fortified wheat. As the temperature and the proportion of added wheat increased, the total Zn content in the extrudates increased. The bioaccessibility of Zn also increased with increasing extrusion temperature, with no statistical difference in values for most samples due to the addition of different proportions of fortified wheat. Total Se content increased with increasing proportion of fortified wheat and decreased with increasing extrusion temperature. There was no significant difference in the bioaccessibility of Se by increasing the extrusion temperature and the proportion of enriched wheat.

Keywords: extrusion, fortified wheat, total starch content, starch damage, bioaccessibility

Comparative study on prebiotic activities of various dietary fibers on distinct probiotics

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Summary

More and more up-to-date studies underpin the relevance of versatile dietary fibers in terms of optimizing balance of intestinal microflora and promoting gut's health, as well as overall human health. Most of the available research results stated that prebiotic materials facilitate the activity and the reproduction of probiotic bacteria. At the first stage of the study inulin was used as model prebiotic, and its impact on activity of distinctive probiotics (*Bifidobacterium lactis*, *Bifidobacterium breve*, *Lactobacillus acidophilus*, *Lactobacillus rhamnosus*) has been assessed. The actual behavior of probiotics can properly be predicted by the application of artificial digestion models, thus the prebiotic effect of several potential food components (chicory, inulin, fructo-oligosaccharides, resistant starch, dahlia, Ganoderma, fenugreek) were investigated. The simplified model includes sterile flasks with enzyme at 37 °C and pH adjusted to 6.8 in anaerobic conditions. The probiotic strains, mentioned above, were added to the model system. The change in the composition of the microbial material was determined by applying a plating technique. The results show that inuline of synergy type and inuline of chicory root exhibited the most outstanding prebiotic activities regarding all the 4 investigated probiotic strains. Resistant starch variants may also contribute to the enhancement of the prebiotic characteristics. Interestingly application of combinations of resistant starches and inuline led to enhanced prebiotic activities in cases of *Bifidobacterium lactis*, *Bifidobacterium breve*. The investigated honey and biscuit matrices tend to reduce the observed prebiotic activity of both fructans and Ganoderma. Fenugreek has not displayed marked prebiotic effect on any of the examined strains. Apparently there is a great potential in the food industry for the application of blends of distinctive prebiotics.

Keywords: prebiotics, probiotics, in vitro model systems, microbial activity,

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Application of *Staphylococcus carnosus* for nitrate reduction in production of semi-durable sausages

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Summary

The aim of this work was to determine the potential of nitrate-reducing starter culture for nitrate reduction in the production of semi-durable sausages and to evaluate its effects on pH, color, water activity, residual nitrate and nitrite content, oxidative stability, and sensory properties of the final product. Five batches of sausages were prepared: one control batch with 100 mg/kg sodium nitrite and four experimental batches with 100 mg/kg sodium nitrate and starter culture *Staphylococcus carnosus* (10^9 /kg). Experimental batches were further divided into sub-batches that were incubated at 30°C and 40°C each for 1.5 and 3 hours, respectively. Finally, all batches were heat treated at 80°C until 72°C was reached in the middle of the product. No significant difference between experimental treatments in pH, color, water activity, or oxidative stability was observed. It was found that residual nitrite content increased and residual nitrate content decreased with increasing temperature and duration of incubation. Residual nitrite content was significantly lower in sausages of experimental treatments than in control treatment. The sensory properties of sausages in treatment with lower temperature and shorter incubation period were not significantly different from those of the control treatment and are considered suitable for the production of semi-durable sausages. Based on the results, addition of *Staphylococcus carnosus* for nitrate reduction during incubation in production of semi-durable sausages proved to be feasible and effective.

Keywords: semi-durable sausages, nitrites, nitrates, starter culture, sensory analysis

Influence of flash thermal treatment on volatile compounds of virgin olive oil

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Summary

The nutritional quality and sensory properties of virgin olive oils (VOO) are strongly influenced by a complex endogenous enzyme system. The activity of these enzymes can be influenced by the process conditions during oil production. The aim of the present study was to determine the influence of flash thermal treatment (FTT) on lipoxygenase activity and on the formation of volatile compounds. Olive fruits from Istarska bjelica and Levantinka were used for this experiment. FTT was performed as a pretreatment to malaxation by rapid cooling or heating from 15 to 40 °C. Lipoxygenase was isolated from the olive paste after malaxation, and its activity was determined by measuring the concentration of hydroperoxides formed by the HPLC method. The volatile compounds of VOOs were analyzed by SPME - GC/MS. The results show that lipoxygenase activity is cultivar dependent, with Levantinka having significantly higher activity. Consequently, Levantinka VOOs have significantly higher concentrations of lipoxygenase pathway volatile compounds. FTT had no significant effect on lipoxygenase activity, although the trend of decreasing activity with increasing temperature was observed. However, volatile compounds of the produced oils were temperature dependent, and their concentration increases from 15 °C to 20 °C for Levantinka and to 25 °C for Istarska bjelica. Thereafter, it gradually decreases with increasing temperature. The concentration of secondary oxidation products was also determined to define the influence of the temperature on the oxidation processes. Neither the cultivar nor the FTT had any effect on the oxidation products.

Keywords: flash thermal treatment, lipoxygenase, virgin olive oil, volatile compounds

Estimation of forage value of grasslands of the Dinaric Mountains based on geospatial data collection

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Summary

The grasslands of Dinaric Mountains have been traditionally used as pastures during the summer months. After decades of abandonment, livestock production on grasslands started to restore but for optimal grassland management and nature protection on a wide scale, it is essential to have digital data on their productivity and feed value. This study aimed to determine the aboveground plant biomass (AGB) and chemical composition of grasslands at 57 locations on the Dinaric Mountains. Between 2020 and 2022, samples of plant material have been collected at different locations of the study area – 26 samples were taken in northern parts near Dinaric peak, 23 in areas near Troglav peak, and eight on Kamešnica mountain. On altitudes lower than 600 m 28 grasslands were sampled whereas 29 were sampled on altitudes over 600 m. Crude protein content, neutral detergent fiber (NDF) content, and acid detergent fiber (ADF) content were determined by analytical chemical analysis. AGB varied between 0.3 t/ha and 5.58 t/ha. NDF content varied between 459.34 g/kg DM and 782.8 g/kg DM (average value: 640.3 g/kg DM) and it was observed that it tended to increase during the growing season. ADF content varied between 291.49 g/kg DM and 507.5 g/kg DM (average value: 395.89 g/kg DM). Crude protein content varied from 36.7 to 188.3 g/kg DM. Statistical analysis showed that the AGB of higher-altitude grasslands was significantly higher than that of lower-altitude grasslands. NDF content was higher on higher-altitudes of Dinaric and Troglav Mountain but not Kamešnica Mountain. Collected geospatial data may be used for the development of a pastoral map for the planning of sustainable grassland resource management on the Dinaric Mountains.

Keywords: productivity, Nature Park Dinaric, karst pastures, ADF/NDF, protein

Acknowledgments: This study was conducted as a part of the project „Dinaric back to LIFE – Management planning and restoration of Dinaric dry grasslands to save biodiversity and support sustainable development“ (LIFE 18 NAT/HR/000847). The project is co-funded by EU and the Environmental Protection and Energy Efficiency Fund.

Sensory properties of semi-hard goat cheese at different ripening times in oil

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Summary

The aim of this study was to investigate the influence of ripening of semi-hard goat cheese in oil (mixture of extra virgin olive oil from Mljet and refined sunflower oil; 50:50) on its sensory properties. Five batches of cheeses were produced, and the cheeses of the same batch were divided into three groups: 1) ripening in air (control group), 2) ripening in oil after 10 days of ripening in air, 3) ripening in oil after 20 days of ripening in air. After 60 days of ripening, the descriptive sensory analysis of cheeses was carried out by the panel of six trained expert assessors. Both ripening in oil and its duration had a significant ($p < 0.05$) influence on the sensory properties of the cheese. Compared to the control group, the cheeses matured in oil had a significantly ($p < 0.05$) higher intensity of an oily odour and flavour, greater elasticity and stickiness determined by fingers but a thinner rind, lower intensity of the cross-sectional colour as well as less crumbliness in the mouth. Overall, ripening in oil improved the thickness of the rind, taste, texture, and cross-sectional properties of the cheese. Cheeses with longer ripening time in air before immersion in oil (group 3) were characterized by higher likeability of appearance, cross-section, odour, and taste compared to the cheeses with shorter ripening time in air before immersion in oil (group 2). It can be concluded that a shorter ripening in oil had a greater positive influence on the sensory properties of the cheese.

Keywords: goat milk cheese, semi-hard cheese, descriptive sensory analysis, ripening, oil

Content of fatty acids in intramuscular fat and subcutaneous backfat in Black Slavonian pigs

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Summary

The aim of this research was to determine the composition of fatty acids in intramuscular (IM) fat and subcutaneous backfat in Black Slavonian pigs raised in free-range conditions. A total of 60 pigs were divided into two groups of 30 animals each, of which one group was fed with acorns in the final period of fattening, and the other group was fed with standard diet. Fattening period lasted from 110 to 145 kg of live weight. The determined profile of fatty acids showed a reduced proportion of saturated fatty acids (SFA) and an increased proportion of monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids in intramuscular fat and subcutaneous back adipose tissue in pigs fed acorn compared to pigs fed with standard diet. Also, the ratio of n:3/n:6 PUFA in the fat of pigs fed with acorns was lower compared to pigs fed diets without acorns. The above results indicate a beneficial effect of adding acorns to the diets of Black Slavonian pigs on the profile of fatty acids in intramuscular fat and backfat tissue.

Keywords: Black Slavonian pig, fatty acids, intramuscular fat, backfat

Effect of dry ageing duration on the physical and sensory properties of beef burgers

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Summary

Dry ageing is mainly used to improve the tenderness and flavour of the meat. It is normally used as a whole carcass ageing for the production of costly cuts where the neck portion is considered a by-product. Therefore, the aim of this study was to determine the effect of dry ageing duration on the physical and sensory properties of beef neck burgers. Neck meat from castrated male Angus cattle weighing 600 kg and aged 22 months was used for the study. The neck samples (N = 12) were hung in a dry ageing chamber for 7 days (group Z-7) and 21 days (group Z-21) at a temperature of $2\text{ °C} \pm 1\text{ °C}$ and a relative humidity of $77\% \pm 3\%$. After ageing period, burgers were prepared from ground neck meat (73.9%) and fat (24.6%) with the addition of 1.2% sea salt and 0.3% ground black pepper. The burgers were then heat treated in a steam convection oven at a temperature of 230 °C to an internal temperature of 64 °C . After heat treatment, the burgers were reweighed to calculate the cooking loss and their diameter was measured in two perpendicular dimensions to calculate the reduction in surface area. The burgers were then left at room temperature for 7 minutes, quartered and stored at 40 °C until sensory analysis. The quantitative descriptive analysis and likeability test were used with nine trained sensory experts in separate booths in the sensory laboratory. A significantly lower cooking loss was observed in group Z-21 (26.52%) compared to group Z-7 (31.31%). However, no significant difference in surface area reduction after cooking was observed. Likewise, the descriptive sensory and liking traits of the burgers were not significantly different between groups Z-7 and Z-21. In view of this, it is contemplated that a shorter ageing period of beef necks could be used in burger making.

Keywords: beef, burger, sensory analysis, cooking loss, surface reduction

Control of food supplements and food with hemp - analytics, legislation and impact on health

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Summary

In recent years, an increasing number of products (food supplements, food and cosmetics) containing hemp can be found on the market of the Republic of Croatia. Particularly noteworthy are the positive effects of the active substance CBD (cannabidiol). CBD is not regulated by law at the EU and Croatian level in food and food supplements. In cosmetics, the requirements for restrictions and prohibitions are clearly defined by law, but there is a well-founded fear that certain products are declared as care products, but are used for other purposes, such as oral and topical application for pain relief, which are not allowed. The aim of this work is to examine food supplements and foods containing hemp in their composition for CBD and THC analysis with reference to the product label. At the same time, compliance with the currently valid legal framework - Commission Regulation (EU) 2022/1393 of 11 August 2022 amending Regulation (EC) No. 1881/2006 regarding the maximum permitted amounts of delta-9-tetrahydrocannabinol (Δ^9 -THC) in hemp seeds and derived products is tested. In the most recent analyses conducted, it was found that 38% of the samples contained THC and CBD, which is not in accordance with the legal framework. Based on this, the monitoring of this type of products on the market should be strengthened.

Keywords: hemp products, CBD, THC, legislative framework, market of the Republic of Croatia

Food fortification – milk, vitamin D and impact on health

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Summary

The importance of vitamin D has been scientifically and professionally confirmed by numerous studies over the years. The effects on the bones and joints of children and adults, as well as on immunity, show that a daily intake is indisputable. Vitamin D is synthesized in our skin under the influence of UV rays. In winter, due to the lack of solar energy, it is necessary to supplement it. Most people use food supplements with vitamin D. However, in some European countries, foods (mushrooms, bread) are fortified with micronutrients, including vitamin D. Milk is one of the products recommended for daily consumption based on the food pyramid, especially for children and adolescents. As part of the EU School Scheme project, milk and/or milk meals are distributed once a week to children in primary and secondary schools. In this work, the vitamin D content in fortified milk and commercial dairy products was analyzed. HPLC method with a DA detector was used to analyze the samples, LOD is 3 µg/l and LOQ is 10 µg/l. Samples were previously prepared by direct extraction with ethanol and hexane, with possible addition of lipase for higher fat samples. The results for vitamin D in fortified milk were compared with the declared values and the recommended daily intake established in the EU regulatory framework. It was found that the analyzed amounts were in agreement with the declared amounts in 70% of the samples. The remaining values determined are within the tolerance limits according to the “Guidance document for competent authorities, tolerances for the control of compliance of nutrient values declared on a label with EU legislation”, EU Commission, Dec. 2012. In view of the above, it is justified to obtain a sufficient intake of fortified milk for the recommended daily dose of vitamin D.

Keywords: analytics, daily intake, food fortification, impact on health, milk, vitamin D

Multiple mycotoxins detected in corn harvested in 2022 from Southeast Europe

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Summary

The aim of the present study was to screen corn samples from different regions of Southeast Europe. The corn samples from Serbia, Bosnia and Croatia were delivered between September and November 2022, and 210 samples were analyzed for multiple mycotoxins. The samples were analyzed by LC-MS/MS triple quadrupole using a multi-mycotoxin method to quantify all mycotoxins present. These included aflatoxins AfB1, AfB2, AfG1 and AfG2, α - and β -zearalenol, zearalanone, zearalenone, diacetoxyscirpenol, HT-2, T-2, 3-acetyl-deoxynivalenol, 15-acetyl-deoxynivalenol, deoxynivalenol (DON), nivalenol, fumonisins FB1, FB2 and FB3, fusaric acid (FA), moniliformin (MON), ochratoxin A (OTA), beauvericin (BEA), and enniatin A1, B and B1. Of the samples, 7% were not contaminated, while 93% contained one or more than one mycotoxin. FA was detected in 83%, FB1 in 79%, FB2 in 68% and FB3 in 53% of the samples. AFB1 was detected in 41% of the samples with an average of 121 ppb. In addition, DON was detected in 11%, OTA in 13% and MON in 34% of the samples. Among emerging mycotoxins, BEA was detected in 68% of the samples. In conclusion, fumonisins, aflatoxins and BEA were the most prevalent mycotoxins detected. The co-occurrence of more than one mycotoxin can lead to additive or synergistic effects when fed to sensitive animal species (e.g., swine and poultry amongst others). This suggests that technologies to control mixtures of such contaminants in feedstuffs predominantly based on corn are needed to minimize impacts on animal development.

Keywords: mycotoxins, corn, LC-MS/MS

A comparative study on the selected quality properties of frankfurters with wild nutria meat

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Summary

The wild nutria (*Myocastor coypus*) is an invasive species in the Czech Republic, and the catch of this species is increasing every year. In terms of sustainability of food resources, the meat from the hunted animals should be further processed. The aim of the study was to compare a control frankfurter sausage made of lean pork (PLF) with frankfurters to which 33% lean nutria meat (NLF) was added instead of lean pork. Most basic chemical and microbiological parameters were not significantly different between these two groups ($P > 0.05$) after storage in 4 °C for 3 weeks. Some differences were found in physical properties of the products. The results showed that there was a significant difference ($P < 0.05$) between all colour parameters (L^* , a^* , b^*) on cross-section of the frankfurters. Surprisingly, a significant difference was found between their textural properties. Unheated NLF sausages were softer ($P < 0.05$) and after heat treatment they became harder than PLF sausages ($P < 0.05$). Contrary to our expectations, the sensory analysis of frankfurters with trained panelists showed no significant differences ($P > 0.05$) between PLF and NLF. In conclusion it can be stated that the substitution of pork with lean nutria meat in this product does not degrade its sensory properties. The finished product is microbiologically safe and can offer an interesting variety to the consumers.

Keywords: meat products, game, colour, sensory analysis

Applicability of near-infrared spectroscopy in the analysis of macronutrient contents of grape pomace and grape pomace mixtures

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Summary

Grape pomace is a by-product of the wine making process which is usually produced in great tonnages after wine production. Its large quantity represents a great environmental risk so the researchers are looking for ways to re-use the pomace in different processes such as composting, animal feed, polyphenol extraction or seed oil production. Among the possible uses of grape pomace is its use in food products. Up till now, there are examples of its use in food products, but mostly as extracts. However, it is important to explore its use “as is” in food products, as well as the possibilities of its macronutrient analysis using fast, non-destructive techniques such as near infrared spectroscopy (NIRs). Therefore, the aim of this paper was to produce mixtures of grape pomace with different contents of macronutrients and record the NIR spectra of the produced mixtures. Produced mixtures differed in the carbohydrate, fat, sugar, fiber and salt content and the differences in the macronutrient content were connected to the NIR spectra of the samples using multivariate analysis – principal component analysis (PCA), partial least squares (PLS) regression and artificial neural networks (ANN). The results showed good applicability of PCA analysis and ANNs in differentiating between macronutrient contents of grape pomace mixtures. However, PLS method showed very low R^2 values, as well as high root mean squared error (RMSE) values, making it not suitable for the use in pomace analysis.

Keywords: grape pomace, food waste, near infrared spectroscopy, macronutrients, multivariate analysis

Acknowledgment: The work was supported by the European Union through the European regional development fund, Competitiveness and Cohesion 2014–2020 (KK.01.1.1.07.0007.)

Carcass traits and meat colour of adult Cres sheep

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Summary

The Cres sheep is one of the nine indigenous Croatian sheep breeds with a population of about 15,000 animals, whose breeding area consists of the islands of Cres and Lošinj. It belongs to the group of breeds with combined production characteristics. Since the potential value of the meat for processing depends on the characteristics of the sheep carcass and the quality of the meat, the aim of this work was to determine the morphometric characteristics of the carcass and the colour of the meat of the culled Cres sheep ewes. The length of the carcass and its halves, the width and depth of the chest, the width of the pelvis, the circumference of the thighs and the length of the hind leg, as well as the thickness of the subcutaneous fat on the loins were measured on a total of 10 carcasses of Cres sheep slaughtered between 5 and 7 years of age. The colour of the meat was determined 24 hours after slaughter at the *musculus longissimus dorsi* (MLD) between the 7th and 8th rib. The average carcass weight of Cres sheep was 19.6 kg, carcass length was 84.5 cm, and hind limb length was 30.1 cm. Carcasses having a good conformation present a smaller trunk and shorter hind limbs (showing extremely high correlation coefficients between carcass length and hind limb length). In Cres sheep, a moderate positive correlation ($r = 0.58$, $P < 0.001$) was found between body length and hind limb length. From the above data, it can be concluded that the carcass of Cres sheep is quite light and spindle-shaped with an average conformation, which is otherwise a characteristic of typical Mediterranean sheep breeds. The determined colour indicators ($L^* = 42.88$, $a^* = 25.77$, $b^* = 10.16$) indicate that the meat of Cres sheep has a bright red colour and as such is of good quality, i.e. acceptable for processing into various meat products: semi-durable sausages, cured meat products, pâtés and the like.

Keywords: indigenous breed, culled sheep, carcass morphometry, meat colour

Changes in butyric acid bacteria spore concentration during alfalfa silage aeration

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Summary

The role of air diffusion during storage and feed-out and the part of aerobic deterioration has been shown to favour butyric acid bacteria (BAB) growth indirectly and to determine the presence of high concentrations of BAB spores in tank milk. The outgrowth of BAB causes late-blowing and off-flavours in cheeses. The aim of this study was to test how different concentrations of added inoculated starter culture for silage alfalfa affect forming BAB by the time after the beginning of aeration. The BAB spore concentration was determined by MPN procedure according to Dutch standards. Decimal dilutions of samples were inoculated on sterilized milk supplemented with glucose and lactic acid, heated at 80°C / 5 min, and sealed with paraffin. After four days at 37°C, BAB concentrations were determined. Results show a statistically significant increase in BAB concentration by the time of aeration independently of inoculums concentration. It can be concluded that using starter inoculums for silage cannot prevent forming BAB, and that attention should be paid to preventing the flow of oxygen during the feed-out process.

Keywords: silage, butyric acid bacteria, starter inoculums, time of aeration

Degradation kinetics of maize carotenoids during storage of grain and complete feeds of differing in commercial hybrids

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Summary

The stability of carotenoids is strongly influenced by light, temperature and oxygen, resulting in their degradation during processing and storage of grains alone or as part of complete feeds. In the present study, the kinetics of carotenoid degradation during storage of complete feeds for laying hens differing in 15 maize hybrids was compared to whole grains of the same hybrids. Carotenoids [lutein (L), zeaxanthin (Z), α - and β -cryptoxanthin, α - and β -carotene, and total carotenoids (TC)] were quantified using RP-HPLC in grains and complete feeds after 0, 14, 28, 56, and 84 days of storage at 20 ± 2 °C. Carotenoid degradation followed first-order kinetics, which allowed evaluation of the degradation rate for individual and total carotenoids. During storage, a higher carotenoid loss was observed in complete feeds than in whole grains ($P < 0.001$). As a result, the degradation rate of both individual and total carotenoids was up to three times higher in complete feeds than in grains (0.0032 vs. 0.0012 day⁻¹ for TC). In both grains and complete feeds, L and Z were more stable than the other carotenoids. The hybrid significantly affected the carotenoid degradation rate in both grains and complete feeds ($P < 0.001$), and the ranges for TC were 0.0006 - 0.0021 day⁻¹ in grains and 0.0021 - 0.0046 day⁻¹ in complete feeds. Carotenoid degradation increases when maize is a component of complete feeds, but some hybrids are more stable and therefore retain more carotenoids during storage.

Keywords: carotenoids, degradation rate, storage, maize, complete feed

Acknowledgments: This work has been fully supported by Croatian Science Foundation under the project ColourMaize (IP-2019-04-9063). The work of doctoral student Dora Zurak has been fully supported by the “Young researchers’ career development project – training of doctoral students” of the Croatian Science Foundation.

The effect of caraway (*Carum carvi* L.) on the performance parameters and quality traits of broiler chickens meat

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Summary

The aim was to study the effect of caraway on performance parameters and yield and quality (structure and sensory quality) of the main edible parts of fast-growing broiler chickens. Caraway nonstandard grains and/or grain fragments could be a part of poultry diets with a potential health benefit due to the content of biologically active substances (carvone, limonene). A total of 184 fast-growing (Ross 308) male broilers were included in the experiment. The experiment lasted 35 days. The control group was fed with a diet without caraway addition. The second experimental group was fed with a diet containing 1% *Carum carvi*. Broilers were fed with experimental starter diets until 11th day of age. Chickens were fed with experimental grower diets from 12th day to 35th day of age. At the end of the experiment, the broilers were weighed and slaughtered. The breast and thigh muscles were deboned and weighed, so that yields of the main parts could be determined, as well as the basic chemical composition (dry matter, crude protein, crude fat). The breast and thigh muscles were subjected to a sensory evaluation, while texture was measured in the breast muscle (TIRA test). When evaluating the data, no significant differences ($p > 0.05$) were found in the growth and carcass yields of the main parts of the chickens. The mean live weight at the end of trial was 2.1 kg. The mean carcass yield was 67.6% from which 34.9% was breast meat and 23.6% thigh meat. No differences were found in basic chemical composition, texture and sensory analysis of breast and thigh meat. Based on the results, it can be stated that the inclusion of 1% caraway in the diets of broilers did not affect performance parameters nor the sensory properties of the meat. It can be preliminarily concluded that caraway may be used as feed for broiler chickens. For more general explanations and recommendations for the evaluation of caraway as poultry feed, further tests are needed.

Keywords: fast-growing broilers, carvone, limonene, poultry nutrition



Session

9 Book of Abstracts

Precision Farming and Sustainable Bio-based
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međunarodni
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Towards a Greater Understanding of Environmental and Cost Impacts using Life Cycle Assessment and Techno-economic Analysis - Increasing Circularity Throughout Farms, Factories, and Supply Chains

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Summary

As agricultural and biological systems evolve due to changes in technologies and production practices, it is increasingly important to understand their environmental impacts, and the costs associate with trade-offs. And, as food and ag supply chains move toward circular systems, quantifying these impacts becomes even more relevant especially as choices are often driven by government and industry policy makers. Life Cycle Assessment (LCA) is a tool commonly used to estimate environmental burdens due to options implemented, and can be used to assess an entire supply chain from cradle to grave, or any sub-portion thereof. Techno-economic analysis is a tool used to estimate potential cost impacts of choices in production systems. Both of these tools are part of the Triple Bottom Line, which is an approach used to assess systems and technologies. The third component is the analysis of social impacts, which is often done via Social Life Cycle Assessment. This presentation will provide an overview of all three tools, and will discuss examples of their use for agricultural and biological systems, including farming and factory operations, including biofuel, bioplastic, and bioproduct production. These examples will illustrate the benefits, but also the challenges of using these tools to make decisions. As farm and industrial systems continue to implement circular approaches, using tools to estimate environmental, cost, and social impacts will be increasingly important.

Keywords: sustainability, life cycle assessment, techno-economic analysis, circularity

Miscanthus x giganteus and *Arundo donax* L. as substrates for augmented biogas production

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Summary

The current EU energy policies aim to increase the share of renewable energy sources to at least 27 % of the total energy consumption until 2030. Biogas is considered a renewable energy source traditionally produced from corn silage or organic waste; however, the transition from traditional to sustainable raw materials, such as agricultural energy crops, is strongly encouraged such as *Miscanthus x giganteus* and *Arundo donax* L. as perennial crops that show high potential for cultivation on marginal lands and exhibit positive energy balance. In this research, *M. giganteus* and *A. donax* were tested as substrates for the anaerobic digestion. The process was enhanced by bioaugmentation and the usage of biocarriers. Bioaugmentation implies the addition of desired microorganisms to the reactors, and a novel approach of isolating the targeted bacteria directly from the inoculum and conditioning them for a specific substrate was tested. Additionally, targeted bacteria were immobilized onto perlite, an aluminosilicate material used as a biocarrier, thus creating the bioparticles to be added to the biogas-producing reactors. Different consortia of bacteria optimized respectively for *M. giganteus* and *A. donax* were isolated and identified by MALDI-TOF technology. When added to the bioreactors, the consortia, both in the form of suspension or bioparticles, significantly increased the biogas yield. However, when consortium optimized for *M. giganteus* were added to the bioreactors supplemented with *A. donax* as substrate, no enhancement in biogas yield was noted, suggesting that bacteria conditioned to a specific substrate could significantly upgrade the biogas production.

Keywords: bacteria, bioaugmentation, anaerobic digestion, biocarriers

The research was funded by the OP “Competitiveness and Cohesion” 2014-2020, project KK.01.1.1.07.0078 „Sustainable biogas production by substituting corn silage with agricultural energy crops“, and by the European Commission and BBI consortium via H2020 BBI-DEMO project No. 745012 „GRowing Advanced industrial Crops on marginal lands for bioEfineries - GRACE”.

Changes in artificial algal-bacterial communities at different CO₂ concentrations

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Summary

The aim of this study was to characterize the microalga *Chlorella vulgaris* SAG 211-11b in the presence of plant growth promoting bacteria (PGPB) at different CO₂ concentrations. PGPB can enhance the growth rate of algal cultures and different microorganisms can help us to create production chains for specific molecules. Elevated CO₂ levels can either represent changes in the atmosphere or flue gases with significant amounts of CO₂. Microalgae are good model systems for both scenarios.

The preliminary results have shown the potential of PGPB on the growth *Chlorella vulgaris* SAG 211-11b. There was a significant increase in the cell density of the algae in co-cultures with PGPB compared to axenic cultures. Possible mechanisms include exchange of nutrients, hormones, volatile organic compounds and mitigation of environmental stresses.

Co-cultures of microalgae and PGPB provide an edge over axenic cultures in terms of robustness, longevity and productivity in general. The co-cultivation of PGPB and microalgae has great potential, as these communities provide similar advantages at elevated CO₂ levels. However, further investigations should be made using actual industrial conditions and large scale facilities.

Keywords: Chlorella, plant growth promoting bacteria (PGPB), CO₂

Hydrothermal carbonization of energy crops

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Summary

The conversion of agricultural biomass by using hydrothermal carbonization (HTC) attracted a widespread attention recently. HTC is a thermo-chemical process in which moist biomass is processed in a hot compressed water at subcritical temperatures, which improves the properties of the raw biomass for its subsequent use. The method was invented to derive the greatest advantage from by-products for soil amendment and electricity production. The main products of HTC process are solid (hydro-char) and liquid fractions. The hydro-char, rich in carbon, can be utilized as a fuel, as an alternative for coal thus replacing a fossil fuel, as a feedstock for gasification, as a soil amendment for potential nutrient enrichment, or as an adsorbent or precursor for activated carbon production. On the other hand, the liquid fraction can be used for further extraction with organic solvents, or in anaerobic digestion. Compared to the pyrolysis process, HTC has several advantages which include zero requirements for pre-drying of the feedstock (biomass), low process temperatures (180–350°C) as well as certain environmental benefits.

Moreover, having in mind that different types of biomass have different shares of cellulose, hemicellulose, and lignin, means that the feedstock must be considered as an important process parameter, along with the reaction time and temperature. Interest in agricultural energy crops such as miscanthus, switchgrass etc. has been increasing recently. Hence, this study aims to explore the process of the hydrothermal carbonization of energy crops such as *Miscanthus x giganteus*, *Arundo donax* L., and *Panicum virgatum*.

Keywords: energy crops, hydrothermal carbonization, hydro-char, zero-waste production

CAPTIVATE supports farmers in the implementation of the Common Agricultural Policy's elements of green architecture

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Summary

Green architecture of the Strategic Plan of the Common Agricultural Policy (SP CAP) of the Republic of Croatia 2023-2027 is based on an increased conditionality, new green interventions within the framework of eco-schemes from the pillar I of the CAP and additional targeted rural development interventions from the pillar II, what contributes to green and climate adaptation of Croatian agriculture. Elements of the green architecture support a more ambitious transition to more sustainable farming systems in accordance with the objectives of the European Green Deal and key strategic documents "Farm to Fork" and "Biodiversity Strategy for 2030."

CAPTIVATE project aims to familiarize farmers with the main green architecture components - rules of conditionality, eco-schemes within the first pillar and agro-environmental interventions within the second pillar - and encourage them to apply sustainable and environmentally friendly measures aimed at reducing the negative impact on the climate and the environment in accordance with the main strategies of the new CAP.

As part of the CAPTIVATE project, IPS Konzalting systematically processes the regulations, rules and measures of the CAP oriented towards contributing to the climate and environmental goals of the CAP, linking them with examples of good eco-practices collected from relevant sources at the national and European level. The goal is to familiarize farmers with the available funding mechanisms linked to concrete examples of good environmental practices in order to achieve responsible and efficient implementation of the Common Agricultural Policy measures.

Keywords: CAP, green architecture, eco schemes, eco-practices, CAPTIVATE

CAPTIVATE (CAP Transfer of Information Via Assessment, Training and Extension) received funding from the Erasmus + program of the European Union No. 2021-1-HU01-KA220-VET-000034777.

Possibilities of energy utilization of CBD extraction by-product

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Summary

During the last decade, the global pharmaceutical industry was primarily focused on the cannabinoid extraction (mostly cannabidiol – CBD) from dry hemp (*Cannabis sativa L.*) biomass. Depending on the extraction method used, the first raw cannabinoid extract amounts to average under 10 % of total biomass. The remaining share of 90 % is waste.

The aim of this research was to determine the usability of industrial hemp waste after the ultrasound extraction in ethanol for the energy production *via* direct combustion and pyrolysis. The biomass of industrial hemp, variety Fortuna, was used in the research. Analyses were carried out by using standard methods for determining the content of proximate and ultimate analyses, Ash content was found to be 15,40 %, coke content was 22,12 %, fixed carbon was 8,12 %, whereas volatile substances were found to be 68,81 %. The obtained HHV was 16,60 MJ/kg, while LHV was 15,35 MJ/kg. After the pyrolysis process, the share of produced biochar (50,45 %) and bio-oil (13,70 %) was determined. The composition of biochar was analyzed using the standard methods – moisture content of 2,71 %, ash 29,22 % and coke 37,06 %. The obtained results confirmed the potential of CBD extraction by-product in terms of high energy value and environmentally friendly raw material for energy production. It has proven to be an attractive source of energy that has the potential to partially replace raw materials of fossil origin in energy production.

Keywords: industrial hemp, CBD extraction, energy, pyrolysis, direct combustion

Tracking phenological changes in different crops using unmanned aerial vehicle

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Summary

Sustainable and profitable crop production is often challenged by abiotic and biotic stress. But with advancement of technology in precision agriculture, application of innovative sensors for data acquiring provided rapid and simplified decision-making tool for timely intervention during vegetation period. Aim of this study is to track phenological dynamics of important continental field crops (wheat, barley, corn, soybean, oats) in area of Experiment Station Šašinovec (University of Zagreb Faculty of Agriculture) using high-tech tool such as UAVs. For acquiring needed information, we used DJI Phantom 4 multispectral drone. This type of high-precision drone, equipped with a multispectral camera array with 5 cameras is widely used for gathering precise, plant-level data. During the growing season there were three different times of data collection. Gathered data was processed with computer software Pix4D Fields, an advanced agriculture mapping software. Using the software, we generated different vegetation indices, such as: NDVI, VARI, TGI, SIPI2, LCI and MCARI. The indices analyse how plants reflect light on a near-infrared spectrum. From several possibilities of indices, we chose Normalized difference vegetation index (NDVI) values as it could give distinguishable results between crops and phenological phases. The goal of the research was to explore the added value of the UAV in the test-area which resulted with the possibility for improving crop production. Furthermore, this research will set the foundation for future collaboration between agricultural experts and scientists.

Keywords: precision agriculture, vegetation indices, UAV

Bioeconomy in the new Programming period 2023-2027

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Summary

The Republic of Croatia has entered the new Programming period of the European Union. Until 2027, the agricultural sector will mostly be financed through the measures included in the new Strategic Plan 2023-2027. Also, apart from the Strategic Plan, funds have been made available through numerous measures and programs as an effort to respond to the crisis, particularly in the areas of bioeconomy development and the creation of added value by using the biomass for energy and new sustainable materials. The research was conducted by using the survey method. 1080 farmers were included in the survey, in which the respondents could choose one of the answers offered. The goal of the survey was to determine the needs of farmers for the new period and their readiness for investments that are also necessary for bioeconomy development. The results showed that the greatest need of farmers concerns the financing of basic machinery and that more than half of the respondents did not utilize the measures from the previous period.

Keywords: machinery, Strategic Plan, investments, bioeconomy

Strength properties of material based on oyster mushroom (*Pleurotus ostreatus*) mycelium

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Summary

The main aim of the research was to produce biocomposite based on oyster mushroom (*Pleurotus ostreatus*) mycelium and straw substrate, and to determine the impact of differences in substrate preparation on the strength of the final product. This research was done through two experiments that differed in substrate preparation and methodology in composite production in several steps. More compact and stable composite was obtained in second experiment, and that allowed flexural strength test using hydraulic press; the flexural strength of the obtained composite was 0.224 MPa and the flexural modulus was 0.007 GPa. It can be concluded that the components of the production process affect the final product, as the strength of the biocomposites prepared using different procedures was different. The treatment, conservation and preparation of the substrate has a major impact on the pH and growth and development of the mycelium, as well as the strength of the biocomposite itself. General conclusion is that the production of mycelium-based biocomposites is simple and cheap, and no expensive equipment is needed for the production process, but it is necessary to comply with the specified conditions for successful growth of the mycelium in the substrate. Also, the mycelium-based biocomposite is completely of biological origin, as it is made of a natural matrix and natural reinforcement and the production of such biocomposite is a good alternative to waste from agricultural production.

Keywords: biocomposite, composite, straw substrate, flexural strength

The influence of fermentation on the proportion of lignin in the silage production

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Summary

Lignocellulosic biomass consists of cellulose, hemicellulose and lignin. Decomposition of such materials is difficult since cellulose is strongly cross-linked and protected by lignin, which disables the access to cellulose for enzymes and microbes. Hence, certain pre-treatments are carried out for the purpose of destruction of the lignocellulosic structure.

For the optimum pretreatment, it is important that the process can make all parts of the biomass available for the formation of bioproducts during further processing. Numerous physical, chemical and biological pretreatments are described for the conversion of lignocellulosic materials; most often, they include the addition of acid, which could have various negative effects on the energy production and by-product utilization, especially in the biogas production.

In this research, the biological method of anaerobic fermentation which occurs during the ensiling process was investigated as a pretreatment of lignin of *Arundo donax* L., which has a common share of lignin of approx. 15 %. The influence of the fermentation on the lignin content was examined in the production of corn silage and ensiled *A. donax* L. In order to determine the proportion of lignin and the influence of the anaerobic fermentation on the investigated samples, the lignin content was determined prior and after the pretreatment. It was determined that the ensiling had a positive effect, and that the lignin content was decreased during the pretreatment.

Keywords: lignin, pretreatment, fermentation, ensiled *A. donax* L., corn silage

FERTIMANURE project - SWOT analysis of bio-based fertilisers created as part of the project

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Summary

The increase in energy prices, as a result of numerous negative events in 2022, caused major changes in all sectors, including agriculture. The shortage and increase in the price of fertilisers indicated the need to ensure the efficiency of their production, which can be achieved by producing fertilisers from animal by-products.

The scientific research project FERTIMANURE deals with this topic. The project is dedicated to the innovative recovery of nutrients from secondary sources for the production of high-added-value fertilisers from animal manure.

As part of the project, with the aim of better understanding and placement of newly produced fertilisers on the market and risk assessment, a SWOT analysis was carried out, which is a simple but powerful tool for evaluating internal (strengths and weaknesses) and external factors (opportunities and threats) of fertilisers. A carefully designed SWOT analysis was used to evaluate important statements between different stakeholders in the EU and different values between stakeholders were observed.

One of the biggest differences is the stakeholders' opinion regarding the impact of the bio-based fertilisers (BBFs) on the reduction of negative impacts on the environment and on the contribution to sustainability. In Germany, stakeholders consider this to be one of the greatest strengths of newly produced fertilisers, while in Italy and Croatia this is not the case. Furthermore, the lack of appropriate legislation at the national and regional levels is considered a major weakness in Croatia and Germany. The European Commission's efforts to research sustainability in the field of application of regulations on fertilisers stand out as the biggest opportunity for Croatian and Italian users. On the other hand, the differences and incompatibility of certification across the EU represent a major threat to the fertiliser production according to stakeholders.

Keywords: FERTIMANURE project, BBF, SWOT analysis, risk assessment, stakeholders

FERTIMANURE has received funding from the EU Horizon 2020., Research and Innovation Programme under grant agreement No. 862849.

Biogas production from corn stalk as a by-product of ensiling the corn cob mix

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Summary

Growing concerns about the current energy crisis and the impact of fossil energy on the environment has recently accelerated the development of sustainable and green energy on a global scale. Following the above, the EC has published the REPowerEU (COM (2022) 109) in May 2022 which encourages the increase of biogas production in the EU member states by 20 % by 2030. Sustainable energy production is mandatory from the entry into force of the EU regulative which defined the goals for reducing the GHG emissions. For the sustainable biogas production, a potential problem arises when using non-sustainable raw material; namely, currently the most commonly used raw material in the biogas production is corn silage of the whole plant. It is well known that corn is used for food and feed, which means that the competition has been created between the production of energy and food/feed from the same crop. Therefore, the aim of this research was to analyse the possibility of using corn stalk by-products in the production of ensiled high-moisture corn cob for livestock feed. The research has shown that the biogas potential of corn silage without cobs (CSWC) was assessed. The interpretation of the data has shown the possibility of using CSWC (100-120 NL/g DM) as a co-substrate in the biogas production, which could lead to reducing the aforementioned competition between energy and food/feed production while ensuring a sustainable biogas production.

Keywords: sustainable biogas production, corn, silage

The research was funded by the OP “Competitiveness and Cohesion” 2014-2020, project KK.01.1.1.07.0078 „Sustainable biogas production by substituting corn silage with agricultural energy crops“.

FairShare project – e-OPG digital platform for farmers

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Summary

The overall goal of the FAIRshare project is to ensure that agricultural advisors and their organizations effectively use digital tools and services to support more productive and sustainable agriculture. The project is co-financed by the EU Horizon 2020 program and gathers 36 partners from different European regions (North-East, South-East, West and Central Europe) that belong to the academic community, research and educational institutions and the agricultural and entrepreneurial sectors. One of the main objectives of the project is to provide an inventory of existing digital tools and support services across the EU and create a comprehensive online catalog containing at least 200 digital tools that farmers can access and use. As part of the FAIRshare project, the Croatian version of the digital platform called “e-OPG” was developed, which is based on the Austrian model “My Farm, My Way”. The e-OPG platform contains important information for farmers in Croatia - including an overview of open tenders, innovative and sustainable agricultural projects and farms in Croatia and the EU, agricultural legislation, success stories of Croatian farmers, as well as a calendar overview of activities in the agricultural sector. Furthermore, the platform offers an overview of a wide range of applications developed as part of the FAIRshare project and ensures quality transfer of knowledge through numerous professional articles and webinars.

The goal of the newly established platform is to facilitate innovation, networking, and education, to create a space where innovative agricultural farms and sustainable agricultural business models are an inspiration for other farmers, and to ensure the exchange of relevant information and knowledge towards farmers, but also among agricultural advisors in Croatia.

Keywords: Horizon 2020, FAIRshare project, digital platform, e-OPG, agriculture

FAIRshare received funding from the European Union’s Horizon 2020 research and innovation program under Grant Agreement No. 818488.

Development of an automatic monitoring system for pear leaf blister moth and its damage using artificial neural networks

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Summary

The effectiveness of pest control depends on monitoring methods, most of which are outdated and irrelevant. For example, the pear leaf blister moth (*Leucoptera malifoliella* (O. Costa, 1836)) is an important apple pest whose larvae damage leaves. Its monitoring is done with pheromone traps, which are time-consuming to inspect and may result in delayed pest control. The most effective time for its control is an early stage of damage occurrence (mines), which is difficult to detect with classical monitoring techniques. Artificial neural networks (ANN) have recently been used for pest monitoring. Therefore, the aim of this study was to develop automatic systems for monitoring pear leaf blister moth and its damage using ANNs. The two ANNs were trained to detect the pear leaf blister moth and its damage using photographs collected in three apple orchards in Zagreb County. The model was developed for use in two prototype smart monitoring devices (pest monitoring device and vegetation monitoring device). The model was tested and the results were processed using a confusion matrix. The developed models showed high accuracy and precision in detecting pear leaf blister moth and its damage, while the monitoring devices showed good functionality in apple orchards. The developed system is effective for automatic monitoring of pear leaf blister moth and its damage, and therefore can significantly improve pest control and apple production in general.

Keywords: *Leucoptera malifoliella* (O. Costa, 1836), precise agriculture, pest monitoring device, vegetation monitoring device, pest control

Attitude of stakeholders regarding the application of Nutri-2-Cycle project priority technologies in Croatia

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Summary

The overall aim of the NUTRI-2-CYCLE research and development project is to assess 16 innovative technologies that ensure a closed nutrient cycle and implementing optimized management systems.

One of the tasks of the project is the implementation of research among relevant stakeholders - farmers, the academic community, agricultural advisors on the implementation and application of technologies of the Nutri-2-Cycle project in Croatia. The objectives of the research include analysis of the innovative technologies' transferability and applicability to other regions and farm types, and it was conducted through individual and group interviews.

Research focuses on 3 main pillars: crop production, livestock production and agro-processing.

From the conducted interviews it can be concluded that the offered advantages of innovative technologies refer to compliance with legislation, better management of nutrients, economic and environmental benefits, practicality in application, while the offered possible obstacles include economic reasons, legislation, necessary knowledge and training for application. Most of the respondents have heard about the presented innovative technologies, but for some they think that they are not applicable in Croatia. The issue of legislation and knowledge and training stand out as one of the main obstacles, while the environmental benefit represents the main advantage of innovative technologies.

As the final conclusion, concerning the short-term (until 2025) and medium-term applicability (until 2030), the majority of respondents believe that most technologies can be applied until 2030.

Keywords: innovative technologies, NUTRI-2-CYCLE, applicability, legislation

NUTRI2CYCLE has received funding from the EU Horizon 2020. Research and Innovation Programme under grant agreement No. 773682



Session **10** **Book of Abstracts**
Plant Protection

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Fitomedicina

Inhibitory effect of pawpaw (*Asimina triloba* L.) extracts on bacteria from genera *Pseudomonas* and *Escherichia*

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Summary

Bacterioses are a significant global problem in plant production. Controlling phytopathogenic bacteria is complex and mostly preventive. Most antibacterial active compounds are copper-based, which are becoming limited in agriculture due to ecological restrictions and the development of resistance in bacterial pathogens. The use of ecologically acceptable natural compounds (plant extracts, essential oils) remains an alternative and insufficiently-considered aspect of plant pathogens, but also animal pathogens contaminating surfaces of plant products. It is known that pawpaw (*Asimina triloba* L.) preparations have an antimicrobial effect on certain pathogens in medicine and veterinary medicine, and this paper tests their effect on bacteria from the genera *Pseudomonas* and *Escherichia*, present on plants.

The antibacterial activity of different aqueous extracts of pawpaw tissues was tested *in vitro* against *Pseudomonas* tomato and *Escherichia coli*. A bactericidal effect was found against the *P. tomato* (the strongest with the seed extract: 92.6% reduction in bacterial colonies compared to the control), while the effect on the *E. coli* was bacteriostatic (highest with the leaf extract: 34% smaller area of colonies in comparison to the control). In the case of the *P. tomato*, a bacteriostatic effect was also detected (highest with the twig extract: 92.1% smaller area of colonies compared to the control).

The results of the conducted research indicate the perspective of the pawpaw tissue extracts as an agents for combating plant pathogenic and plant contaminating bacteria.

Keywords: phytomedicine, plant pathogenic bacteria, control, food quality, prevention

First record of *Penthaleus major* (Acari: *Penthaleidae*) in the Republic of Croatia

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Summary

The blue oat mite *Penthaleus major* (Dugès, 1837) is an important pest on many agricultural crops. The mites penetrate the epidermal cells of the host plant what results in silvery to grey patches on the leaf surface and at high levels of attack over several days, the tips of the leaves turn brown, and the plant may die. *P. major* feeds primarily on Poaceae (oats, phalaris, ryegrasses, wheat) but also on plants like capeweed, lettuce, clover and lucerne. In Europe, the blue oat mite was found in France, the Netherlands, Norway, Germany, England, Iceland, Switzerland, Poland and Hungary. The first appearance of this pest in the Republic of Croatia, at the locality Kakma, in Zadar County, was suspected in December 2021. As it is an unknown species in the Republic of Croatia until recent time, the aim of this research was to identify the species which could become dangerous pest in our climate. Identification was carried out using molecular diagnostics according to Kontschán et al. (2018). Upon multiplication of molecular markers 18S rRNA, cox 1 and ITS2 by polymerase chain reaction, samples were sequenced and bioinformatic analysis confirmed *P. major*. In the Republic of Croatia this species has caused damage to Swiss chard, so it is important to test other host plants. The use of pesticides was and still is the most common option for controlling this pest. In this moment it is important to give priority and to investigate efficacy of non-chemical pest control measures.

Keywords: winter grain mite, Swiss chard, economic pest, molecular diagnostics, Zadar county

Angelica archangelica L. hydrolate – the effect on some cultivated and weed species

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Summary

Many studies are investigating the potential environmentally acceptable herbicides in order to reduce the harmful effects of agriculture. Since *Angelica archangelica* L. expresses many biological activities, the aim was to determine the effect of its hydrolate towards some cultivated (soybean and sunflower) and weed species (*Amaranthus blitoides* and *Chenopodium hybridum*). The seeds (25 of each species in 4 replicates) were put in Petri dishes on filter paper and treated with 10, 20, 50 or 100% *A. archangelica* hydrolate, i.e. the distilled water in the controls. The seeds were kept in a climate chamber during 8/16 hour photoperiod, at 22°C/20°C and 60±1% humidity. The germination was measured daily, while the shoot and root lengths were measured at the end of the experiment. Germination percentage was greater in soybean (15-100%), and lower in sunflower (17-26%), including the controls. For *C. hybridum* the germination was 28-78%, i.e. 19-84% for *A. blitoides*, and all the variants had lower germination than the controls. The shoot and root length in soybean were higher in all variants than in the control. The weeds expressed significantly lower shoot and root length than it was observed in the controls. Further research on other cultivated and weed species in field conditions is needed to fully estimate the potential of this hydrolate.

Keywords: *Amaranthus blitoides*, *Chenopodium hybridum*, soybean, sunflower

Emergence dynamics of *Echinochloa crus-galli* and *Setaria pumila* from an artificial soil seed bank

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Summary

Echinochloa crus-galli and *Setaria pumila* are the most common annual grass weeds in summer crops. In the early growth stage, the two species are morphologically similar but differ in their sensitivity to various herbicides. To distinguish these species and determine the appropriate timing for control, the dynamics of their emergence were studied. Seeds of the two species were collected in Šašinovečki Lug (45°50'59.6" N 16°09'53.9" E) in autumn 2019 and 2021 and buried in weed-free soil at the Experimental Station of Faculty of Agriculture, Maksimir (45°82'62.67"N, 16°02'93.6"E) in November 2019 and 2021. In spring 2020 and 2022, the soil was disturbed to simulate seedbed preparation. Emergence was recorded three times a week. In both years, *S. pumila* emergence started earlier (on the 2nd and 4th days after soil disturbance in 2020 and 2022, respectively). The median (T_{50}) thermal (TT) and hydrothermal time (HTT) to emergence was calculated. The value of T_{50} calculated as TT was 26.7 and 83.7°Cd for *S. pumila* in 2020 and 2022, respectively, whereas higher values were found for *E. crus-galli* (132.02 vs. 187.15°Cd, in 2020 and 2022). The HTT T_{50} for emergence was 10.35 and 34.42 MPa°Cd for *S. pumila* and 46.47 and 115.74 MPa°Cd for *E. crus-galli* in 2020 and 2022, respectively. The results indicate that *S. pumila* is the species with the earliest onset and shortest duration of emergence in the field. On the other hand, *E. crus-galli* has later and longer duration of emergence.

Keywords: barnyardgrass, hydrothermal time, monocot weeds, thermal time, yellow foxtail

Resistant populations of Johnsongrass (*Sorghum halepense* L.) and common ragweed (*Ambrosia artemisiifolia* L.) in Croatia to ALS herbicides

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Summary

The objective of this study was to confirm the resistance of *Sorghum halepense* and *Ambrosia artemisiifolia* to acetolactate synthase herbicides (ALS). Seeds of plants that had survived a ALS herbicide application (R) were collected from soybean and corn fields in fall 2018. A susceptible population of both species that had never been treated with ALS herbicides (S) was also included in the experiment. Bioassays on whole plants were conducted with linearly increasing (R) and linearly decreasing (S) herbicide doses at BBCH 13 and 12, respectively, for *S. halepense* and *A. artemisiifolia*. The bioassays included 9 doses ranging from ½ to 64 times the recommended field dose (x). The experimental design was a completely randomised design with 4 replicates and 5 plants per pot. The logistic curve was used to estimate the herbicide dose required to reduce 50% of the aboveground mass, and then the resistance index ($GR_{50} R/GR_{50} S$) was determined. The results indicate that all herbicides were very effective on the S populations of both species. However, a high level of resistance was found to all herbicides tested: 81.86 for nicosulfuron, 370.78 for foramsulfuron, and 155.47 for imazamox in *S. halepense* and 293.15 for foramsulfuron and 31.7 for tifensulfuron in *A. artemisiifolia*. The detected resistant populations urgently require the long-term incorporation of other weed control methods due to the long viability of the resistant dormant seeds of both species in the soil seed bank.

Keywords: foramsulfuron, tifensulfuron, imazamox, nicosulfuron, maize, soybean

Preliminary results of sweet potato whitefly feeding behaviour on tomato plants using electrical penetration graph

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Summary

Sweet potato whitefly, SPW, (*Bemisia tabaci*) causes extensive damage to vegetables in greenhouses worldwide. The control of insects such as potato whitefly is very difficult, but the study of their feeding behavior can provide adequate information on the factors that disturb the growth and development of these pest. Electrical penetration graph (EPG) is a useful tool to study the feeding habits of sucking insects. The objective of our study was to record and interpret SPW feeding on untreated and treated (2% kaolin clay) tomato plants using the EPG Giga-8dd. Recordings were scheduled for seven hours with eight channels connected to eight plants during each recording. The experiment was repeated for four days. The data were analyzed using the EPG Stylet+a program. The results of EPG recordings showed that the first waveforms appeared when the leaf tissue was pierced, mostly in the third hour after the beginning of harvest. The C wave occurred in all individuals (100%). The E1 waveform was of shorter duration and occurred before the E2 wave. The E2 wave, i.e., the phloem-swallowing wave or feeding, occurred after the E1 wave and lasted much longer, approximately 27 minutes. Wave G (xylem feeding) was achieved by 87.5% of individuals on treated plants four times during one recording, and 75% on untreated plants three times per insect. When comparing C-wave values in recordings made on untreated and treated tomato plants, it was found that plant treatment had no effect on leaf tissue probing success. Using this method, it is possible to visualize the feeding behavior of hemipterans in real time. Understanding this process could lead to finding weak points in feeding and better protecting plants from this harmful species.

Keywords: *Bemisia tabaci*, hemipteran insects, electrical penetration graph, insect-plant interactions

Integrated weed control in maize: sustainable solution to control ALS resistant Johnsongrass in Croatia

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Summary

Maize is one of the most important crops in Croatia (300,000 ha) and Johnsongrass is the predominant perennial weed species in some large maize growing areas, which is successfully controlled with ALS herbicides. However, in the Republic of Croatia, target-site resistance of Johnsongrass to ALS inhibitors (MoA HRAC group 2) was confirmed in 2017. The main objective of the work was to develop sustainable control methods against ALS resistant Johnsongrass populations in maize. Multi-year field trials (2018-2022) were established in Novaki Oborovski (45.723551, 16.22972) with the goal of controlling ALS -resistant Johnsongrass. In the five-year trials, different weed control measures were integrated (ploughing, crop rotations, inter-row tillage, use of different MoA) and evaluated how these measures contributed to the reduction of Johnsongrass population in the field and long-term rhizome mass in the soil. The results clearly showed that the integration of different weed control strategies significantly contributed to the reduction of ALS -resistant Johnsongrass (Plots 3-4-5; 2018 no.of shoot/m²: 139-169-162, rhizome mass g/m²: 727-1264-1400, 2022 no.of shoot/m²: 2-0-3, rhizome mass g/m²: 0). The use of different MoAs significantly reduced ALS-resistant Johnsongrass even in monocultures. The best control of ALS -resistant Johnsongrass was shown when the herbicide Adengo was followed by the herbicide Laudis (MoA HRAC group 27+ 2 fb MoA group 27) in combination with inter-row cultivation (efficacy average 91.9%). Integrated weed management are the best solution in controlling resistant Johnsongrass, but also ensure long-term sustainable weed control.

Keywords: IWM, Sustainability, ALS inhibitors, Johnsongrass, weed resistance

Inhibitory potential of olive leaf extract on plant pathogenic fungus *Verticillium dahliae* Kleb.

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Summary

Olive is one of the most important Mediterranean fruit and oil crops but its sustained cultivation is endangered by various abiotic and biotic stressors, highlighted by plant pathogenic bacteria and fungi causing serious diseases which can even result in dieback of olive orchards. Verticillium wilt, caused by fungus *Verticillium dahliae* is considered the most important fungal disease of olive with increasing incidence in Europe. *V. dahliae* population infecting olive comprises of highly pathogenic “defoliating” (D) type and less pathogenic, but still dangerous, “non-defoliating” (ND) type, with only latter being identified in Croatia recently. Olive leaves, an important byproduct of olive cultivation and also recently recognized as a source of important natural compounds with antibiotic and antimycotic properties, are used for production of olive leaf extract (OLE). The aim of this study was to screen potential of water solutions of commercially available OLE (Magdis, Zagreb, Croatia) to inhibit growth *in vitro* of two *V. dahliae* (ND) isolates, obtained from Spain and Croatia. The fungal isolates were grown on PDA medium amended with water solution of OLE in four concentrations, with water only for control. Growth of *V. dahliae* was inhibited by 2, 2, 12, 45 % (Spanish isolates) and 9, 36, 70, 81 % (Croatian isolates) at OLE concentrations of 5, 50, 500, 5000 mg/L, respectively. Results suggest variable susceptibility of isolates of *V. dahliae* to OLE with high susceptibility of Croatian isolate to even low concentrations of OLE. In addition to growth rate, OLE reduced fungal colony density and microsclerotia formation. These results suggest future prospect of using OLE in novel methods of biological control of Verticillium wilt of olive.

Keywords: *Olea europaea*, leaf extract, Verticillium wilt, biological control, non defoliating

Monitoring of root-knot nematodes in solanaceae vegetable in Bulgaria

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Summary

The root-knot nematodes (*Meloidogyne* spp.) are one of the main yield-limiting factors in the monoculture production of tomatoes and potatoes (*Solanum lycopersicum* L. and *Solanum tuberosum* L.) in greenhouse and field production. The observed climate changes have a wide impact on ecosystems and agrocenoses. The life cycle of many animal and plant species is changing. Globally, the range and the area of natural dispersal potential of several pests is expanding. The aim of the present research was to study the distribution and species composition of root-knot nematodes of genus *Meloidogyne* in solanaceae vegetable crops, as well as their changes.

The research was carried out in the period 2020-2021 in sites located in the region of Southwestern Bulgaria, Sofia, Blagoevgrad and Pazardzhik regions, which is one of the main producers of tomatoes and potatoes in Bulgaria. Soil and plant samples of 25 points were taken during the vegetation of the plants. The species composition of the extracted nematodes was determined by morphological characteristics of females and juvenile stages. The results showed the presence of root-knot nematodes of the genus *Meloidogyne* in 100% of the tested samples from tomato greenhouses and 85% of the field potato samples, which confirms their wide distribution of these pests in the area. The species *Meloidogyne arenaria*, *Meloidogyne incognita* and *Meloidogyne javanica* were identified. Mixed infection (*M. arenaria* + *M. incognita*) was observed in 73.6% of the greenhouse samples. The incidence of *M. arenaria* was higher in the field and in the greenhouses.

The obtained information will enable tomato and potato growers to develop and implement an appropriate control strategy for these species and will help to carry out a comparative analysis of changes in the species composition of root-knot nematodes.

Keywords: root-knot nematodes, tomato, potato

The occurrence and presence of *Botryosphaeria* dieback of olive trees and fruits in Istria

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Summary

Some of the phytopathogenic fungi from the *Botryosphaeriaceae* family can cause diseases on olive trees. The disease symptoms occur in the form of dieback and rotting of fruits, dieback of olive leaves and branches, the occurrence of necrotic lesions and changes in the color of the bark. The presence of several species from this family has been identified on the olive trees so far in Republic of Croatia. In 2021 and 2022, a field survey, and sampling of symptomatic olives was carried out on Istrian County territory. The main goals of the conducted research were: to determine the cause of symptomatic changes in olive trees in field research in Istria, and to carry out morphological and molecular identification of species of phytopathogenic fungi from the family *Botryosphaeriaceae*. The presence of the phytopathogenic fungi of the *Botryosphaeriaceae* family has been detected in 10 out of 25 locations visited. The largest number of samples was collected in the vicinity of Vodnjan and Rovinj, and phytopathogenic fungus were isolated from some of the most important olive varieties in Istria. Phytopathogenic fungi isolated from the infected plant material (15 isolates in total) have been identified on the basis of morphological and molecular characteristics. Molecular diagnostic was made using the polymerase chain reaction (PCR) method and multigenic analysis of the sequences ITS (internal transcribed spacer), TUB2 (beta-tubulin) and EF-1 α (translation elongation factor 1- α). The research results indicated that the most prevalent species was the *Botryosphaeria dothidea*.

Keywords: *Botryosphaeriaceae*, phytopathogenic fungi, isolate, field survey

Methods of insects monitoring in agricultural production

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Summary

Insects are one of the most numerous group of animals on earth. Studying insects is very important for improving environmental quality, reducing pesticide use, increasing crop productivity and controlling production costs. Insects are mostly beneficial, however a small group of insect pests cause economic losses by attacking crops, damaging food, vectoring diseases on cultivated plants etc. This review study presents an outline of methods that entomologists commonly use in monitoring insects in agricultural crops. There are many methods suitable for monitoring insects in different crops and one of the mostly used are visual detection of insects on plants, and by using different types of sticky traps and nets for insect collecting. Monitoring and sampling of insect underground can be done in different ways: soil sampling by the core, digging a pit, using baits and attractants (e.g. germinating seed baits), Berlese funnels, and for the ground insects the most commonly used method is pit-fall traps. Some insects are monitored by olfactory stimulants, so variations of the abovementioned methods like sticky traps are used in combination with pheromone attractants. However, different sources such as light, scents, or food are used to attract and catch insects. Common attractant includes black light, chemical-attractant (pheromones), food-bait, attractive colors or shapes. Today, insects monitoring methods can be very complex and technologically advanced such as infrared sensors, motion detectors, video surveillance and they are used in combination with artificial intelligence (smart traps). The monitoring method depends on many factors such as plant species and development stage, insect species and development stage, insect population size and all this methods are necessary for making pest management decisions.

Keywords: insect traps, attractants, soil sampling, smart surveillance,

The effect of essential oils on mortality of Colorado Potato Beetle (*Leptinotarsa decemlineata* (Say, 1824))

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Summary

The objective of this work is to determine the effectiveness of essential oils on the mortality of adult Colorado potato beetles. In May 2022, adult forms were collected from two locations on untreated fields in Sisačko-moslavačka County. Under laboratory conditions, the efficacy of six essential oils (star anise, basil, eucalyptus, lemon, lavender, and mint) on adult beetle forms was tested using IRAC test method 007. All essential oils tested were found to be very effective, even at low concentrations. Star anise oil showed the greatest efficacy (55 - 100%) at all concentrations applied, while lavender essential oil showed the least efficacy (0 - 20%) at all concentrations applied 24, 48, and 72 hours after the start of the experiment. Considering the problem of resistance of Colorado potato beetle to chemical insecticides, essential oils show good properties and could be an effective substitute for chemical insecticides, but further studies are needed to confirm this.

Keywords: Colorado potato beetle, resistance, star anise, basil

Reliability of capture the olive fly *Bactrocera oleae* (Rossi 1790) on yellow plates using visual data processing techniques

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Summary

Food production is increasingly preoccupied with precision agriculture. One of its solutions are visual data processing technologies - object detection (OD), combined with the development and application of artificial intelligence (AI) models. These technologies as a tool are much more precise than looking with the “naked eye”. The olive fly *Bactrocera oleae* (Rossi 1790) is an economic pest of the olive fruit that appears regularly in the Mediterranean climate, but the capture is not the same every year. Measuring the capture on the yellow plates, creating the flight curve is an extremely time-consuming job for olive growers. OD technologies, with the application of AI, speed up the process of measuring captures. The developed AI model bridges the spatial distance and travel time of the yellow plates. The aim of the paper is to show the development of the OD model for *B. oleae* and its reliability. The AI model was developed in Zadar County on visual samples of images of yellow plates from 6 localities collected from 2020 to 2022. The research was carried out as part of the project: SAN-KK.01.2.1.01.0100 (Smart agriculture network), financed by IRI- ERDF fund. AI model development was carried out using TensorFlow software. The concept used to determine the level of precision bio efficientdet lite4. This is an AI algorithm, and it works by having a separate set of images determine the quality of the model. With it, reliability of up to 95% was achieved. OD technologies, along with the development of AI models, have proven to be applicable in measuring the captures of adults and setting the flight curve of the olive fly. Technology further bridges distance and time to measure adult captures.

Keywords: annotation, artificial intelligence (AI), Mediterranean, object detection (OD), olive fly

Effect of quinazolinones derivatives on greater wax moth (*Galleria mellonella* L.)

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Summary

Quinazolinones are class of fused heterocycles which exhibit insecticidal activity and broad spectrum of biological activities. Ten quinazolinones based Schiff bases, newly synthesized were tested for their insecticidal activities against greater wax moth (*Galleria mellonella* L.). Oral bioassay was conducted on last stage insect larvae, which were fed with the control diet or the diet containing 100 mg of tested compound per kg of diet. Mortality, change in behavior (orientation test), symptoms on cuticle (change in color, lesions and spots), and effects on molting were monitored after 24, 48 and 72 h. The compounds did not cause mortality in insects, or change in insect behavior. Larvae after turning on its back were able to return to upright position till pupation. After 24 h, eight compounds induced changes on insect cuticle. The greatest number of larvae exhibited change in cuticular color after 48 h. Compound SB17 caused brown and black spots on the cuticle in highest number of insects in each monitoring period. After 72 h, all compounds induced molting to pupa (min 3,33% to max 60% larvae pupated), compared to the control where no pupae were observed. Compounds SB3, SB8, SB14, SB21, SB17, SB19 and SB25 induced earlier pupation in min 40% larvae. The highest and statistically significant effect on earlier pupation was observed for compound SB3, which induced molting to pupa in min 60% of larvae after 72 h. Molting starts after juvenile hormone secretion drops in the last larval instar. Compounds SB3, SB8, SB14, SB21, SB17, SB19 and SB25 should be further tested for their activity as insect growth regulators.

Keywords: Schiff bases, insect mortality, insect behavior, cuticular changes, molting

Acknowledgment: Project NSF “Green Technologies in Synthesis of Heterocyclic Compounds” (UIP-2017-05-6593)

Current status of the quarantine pest *Aleurocanthus spiniferus* (Quaintance, 1903) (Hemiptera: Aleyrodidae) in Croatia

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Summary

In Croatia, the species *Aleurocanthus spiniferus* (Quaintance, 1903) was first recorded in 2012 on ornamental seedlings of bitter orange (*Citrus aurantium* L.) in the garden center in Split. After microscopic identification and species confirmation, eradication measures were applied, and it was not recorded until 2018. In September 2018, by carrying out special surveillance of quarantine species of whiteflies from the genus *Aleurocanthus* Quaintance & Baker, 1914, in Vitaljina, *A. spiniferus* was visually detected in a mandarin orchard, and later confirmed by microscopic identification. The aim of this paper is to show the current distribution, harmfulness and phytosanitary status of the species *A. spiniferus* in Croatia after the first positive finding. The species *A. spiniferus* has firmly established itself in southern Croatia. The developmental stages of the pest were found in the mandarin orchard and on wild plants near the orchards. In the following years, the pest spread towards the northwest of Dubrovnik-Neretva County. It has also been confirmed in Split-Dalmatia County. Development stages in a very high population were also found on other different plant species such as *Vitis vinifera* L., *Hedera helix* L., *Rosa canina* L., *Prunus laurocerasus* L., etc. Due to its marked polyphagy, there is a danger of further spread, which represents a high phytosanitary risk for citrus groves in the Neretva valley, vineyards on the island of Hvar and other agricultural crops in Croatia.

Keywords: orange spiny whitefly, damage, host plants, distribution, quarantine pest

In vitro maize selectivity to three phenolic acids that inhibit *Ambrosia artemisiifolia* L. growth

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Summary

Phenolic acids are natural compounds that can inhibit the growth of many weed species. The objective of this study was to test the inhibitory potential of ferulic acid (FA), vanillic acid (VA), and *p*-coumaric acid (PCA) on the selectivity of *Ambrosia artemisiifolia* and maize *in vitro*. The phenolic acids were applied individually to the seeds in doses (10^{-7} mol): 200 (FA, VA, PCA), 400 (FA, VA), and 600 (VA). Treatments were applied to 50 seeds of *Ambrosia artemisiifolia* and 25 maize seeds per Petri dish in three replicates. The inhibitory potential of phenolic acids was estimated from the reduction in germination and early growth parameters 14 and 7 days after treatment of *Ambrosia artemisiifolia* and maize, respectively. The obtained data were subjected to analysis of variance and differences in mean values were tested in R using Tukey's test ($p < 0.05$). PCA showed the greatest overall reduction potential, reducing radicle and hypocotyl length by more than 60%. Higher inhibition was achieved only with 600×10^{-7} VA by reducing germination by 85%. Maize was not significantly affected by the phenolic acid treatments. The highest reduction of radicle was caused by 200×10^{-7} mol PCA and 400×10^{-7} mol FA (16%). Based on the reduction potential against *Ambrosia artemisiifolia* and the selectivity of maize to PCA and VA, the effect of these phenolic acids will be further investigated in combination applications with reduced doses of herbicides used in maize.

Keywords: ferulic acid, *p*-coumaric acid, vanillic acid, allelochemicals, integrated weed management

Effect of water stress on allelopathic potential of petunia (*Petunia hybrida* L.)

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Summary

The aim of the paper was to evaluate the influence of water stress i.e., different net irrigation water on the allelopathic potential of petunia (*Petunia hybrida* L.). Water extracts were prepared from aboveground biomass of petunias grown at different net irrigation water (51.5 mm, 91.5 mm and 132.5 mm) in three concentrations (1%, 2.5% and 5%). The experiment was performed under controlled laboratory conditions in Petri dishes and the allelopathic potential of petunia water extracts was assessed by measuring seed germination, root and shoot length, and fresh weight of radish seedlings. The results of the experiment showed a statistically significant allelopathic effect of petunia water extracts on all the measured parameters. The allelopathic potential largely depended on the extract concentration. Petunia extracts of higher concentrations showed a greater negative effect and reduced germination, shoot and root length, and fresh weight of seedlings up to 79.9%, 93.4%, 83.1% and 69.5%, respectively. Positive effect was determined on shoot length and fresh weight of radish seedlings with 1% concentration extracts. As a rule, water stress i.e., net irrigation water did not have a significant effect on the allelopathic potential of petunia. The exception was recorded with the highest extract concentration on the shoot length of seedlings where extracts from petunias grown at 51.5 mm had greater inhibitory effect compared to extracts from petunias grown at 91.5 and 132.5 mm.

Keywords: water stress, allelopathy, net irrigation water, concentration, radish

Toxic effect of some essential oils on storage pest *Tenebrio molitor* L.

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Summary

Tenebrio molitor L. damages stored flour and flour products, grain, milk powder and meat, while the larvae also damage the packaging. Controlling this pest has become difficult due to the reduction of available active substances and the development of resistance to insecticides. The toxic effect of essential oils of basil, lavender, eucalyptus, star anise and lemongrass was investigated in the work, since some essential oils show insecticidal activity. The fumigant effect of ten concentrations (2, 4, 8, 16, 32, 200, 400, 600, 800 i 1000 µl/l air) of essential oils of basil, lavender, eucalyptus, star anise and lemongrass was tested on yellow mealworm larvae. Ten larvae were placed in a glass container, and filter paper treated with an essential oil was glued to the underside of the lid. Each concentration and control (water) was set up in five replicates. Mortality was recorded every 24 hours for five days. Eucalyptus essential oil showed the best results at concentrations higher than 200 µl/l of air 24 hours after setting up the trial, while essential oils of basil, lavender and star anise had similar effect evident four to five days after the treatment. All five essential oils in concentrations up to 32 µl/l of air showed very low fumigant toxic effect (less than 6 %). The researched essential oils could be used as an alternative and ecological solution in the control of this important pest of stored products.

Keywords: botanicals, fumigants, storage pests, yellow mealworm beetle

Role of Silicon foliar application in defence against jasmine moth in olive groves

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Abstract

The aim of this work was to investigate the influence of the silicon foliar application on the intensity of olive damage caused by the settlement of jasmine moth (*Margaronia (Palpita) unionalis* Hubner) larvae, and the phenolic response of olive leaves.

The field trial on two years old Leccino cv. olives was conducted, as a completely randomized design with two treatments (untreated control (Si-) and silicon treated plants (Si+)), during September and October of 2022. During the trial Si+ plants were foliarly treated five times at intervals of one week. After second treatment the larvae of four jasmine moth were settled on the apical buds of Si- and Si+ olives. Seven days after the last treatment, samples of young and old leaves for phenolic analyses were taken. Damaged leaves, apical buds and the number of pupae per plant were also counted.

The results showed significant differences in the oleuropein concentration between Si- and Si+ leaves (6121 mg/100 g DW and 6914 mg/100 g DW, respectively). Comparing the percentage of damaged leaves and the percentage of damaged apical buds, there was no significant difference between Si- and Si+ olives. A significant difference was shown in the number of pupae per plant, namely, no pupae were found on Si+ plants, while an average of 0.3 pupae per olive was found on Si- plants.

Due to our preliminary results further investigation of the silicon foliar application in protecting olives from the jasmine moth as well as other insects is needed.

Keywords: Leccino, *Margaronia (Palpita) unionalis* Hubner, olives, leaf, oleuropein

Bio-herbicidal potential and chemical analysis of secondary metabolites of field dodder (*Cuscuta campestris* Yunck.)

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Summary

The objective of this study focused on characterizing chemically and evaluating *in vitro* the allelopathic and bioherbicidal potential of secondary metabolites (phenolic compound) extracted from the stem of field dodder (*Cuscuta campestris* Yunck.) in seed germination, early seedling growth (radical and shoot length) and early plant growth tests of *Amaranthus retroflexus* L. and *Portulaca oleracea* L. The combined effects of the stem extract and a reduced dose of the synthetic herbicide metribuzin on the two weed species were also examined. Plant extract contained 18 phenolic compounds and the most abundant phenols were flavonoids: quercetin, (+)-catechin, daidzin, luteolin, and rutin. The seeds of *P. oleracea* were less sensitive than the seeds of *A. retroflexus*. The data obtained in the seed bioassay confirmed the inhibitory effect of plant stem extract on the germination and early growth of *P. oleracea* and *A. retroflexus* seedlings at higher concentrations (0.75% and 1%), and a minor inhibitory effect was also observed in the plant bioassay with this extract. On the other hand, a synergy of *C. campestris* stem extract and the herbicide metribuzin was revealed, as their combination was found to achieve better results in the control of both weed species. Based on obtained data *C. campestris* stem extract could be a potential source of natural herbicide. Also, these findings could be useful in light of increasing practices of integrated weed management that have heightened the interest in reducing herbicide doses.

Keywords: bio-herbicide, plant extract, field dodder

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