FACULTY OF AGROBIOLOGY AND FOOD RESOURCES

FACULTY OF BIOTECHNOLOGY AND FOOD SCIENCES

FACULTY OF HORTICULTURE AND LANDSCAPE ENGINEERING

Scientific Conference of PhD Students of FAFR, FBFS and FHLE SUA in Nitra

with international participation

2021

on occasion of the Science and Technology Week in the Slovak Republic

SLOVAK UNIVERSITY OF AGRICULTURE IN NITRA

TÝŽDEŇ VEDY A TECHNIKY NA SLOVENSKU

10th November 2021
Nitra, Slovak Republic

Slovak University of Agriculture in Nitra

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Proceedings of abstracts

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Preface

In these days, in international media, we are seeing headline after headline focusing on COP26. COP26 is the 2021 United Nations climate change conference, which takes place in Glasgow, Scotland. It is the moment, when countries update their plans for reducing emissions and the nations are working together to reach the agreement how to tackle climate change. Slovak University of Agriculture in Nitra joined to this initiative to protect the climate and planet symbolically, by illuminating the building in green.

Climate has obvious and direct effects on agricultural production. Even though the agriculture contributes substantially to greenhouse gas emission, it has also a unique position to tackle climate change. Tackling climate change will require a widespread shift away from pesticide-dominated agriculture to agroecology. This will lead to resilient, regenerative farming systems that emit less greenhouse gases, improve biodiversity and produce good yields and fair rewards for farmers. To meet the growing consumption needs of the global population will require a successful and environmentally safe application of biotechnology in agriculture. The success of all these issues is depending on the level of our knowledge. But let´s remember

"***Knowledge is not power****; it is only a potential. Applying that knowledge is power. Understanding why and when to apply that knowledge is wisdom*" (Takeda Shingen).

Wisdom of the farmers is developing over a period of time through accumulation of experiences and intimate understanding of the landscape environment. This long-term process underlining the importance of traditional knowledge.

The Scientific Conference of PhD students was organised at the Slovak University of Agriculture in Nitra on occasion of the Science and Technology Week in the Slovak Republic under the auspices of doc. Ing. *Peter Ondrišík*, PhD. – dean of the Faculty of Agrobiology and Food Resources (FAFR), prof. Ing. *Norbert Lukáč*, PhD. – dean of the Faculty of Biotechnology and Food Sciences (FBFS) and prof. Ing. *Dušan Igaz*, PhD. – dean of Faculty of Horticulture and Landscape Engineering (FHLE).

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The main aim of the conference was to provide a platform for knowledge sharing by presenting of research findings and exchange of expertise and experiences. Abstracts in this proceeding were split among the eight areas covered by conference:

- Biotechnology
- Animal production
- Applied and molecular biology
- Nutrition
- Multifunctional agriculture, environment, landscape architecture and rural development
- Plant production
- Technology, quality and safety of raw materials and foodstuffs of animal origin
- Technology, quality and safety of raw materials and foodstuffs of plant origin

We believe that the Scientific Conference of PhD students has encouraged the further advancement through fruitful discussions among students and other participants; and that the scientific programme have contributed to increasing the knowledge, improving the future work, and building new friendships between PhD students from different countries and from the broader spectrum of biological sciences.

doc. Ing. Peter Ondrišík, PhD.
Dean of FAFR

prof. Ing. Norbert Lukáč, PhD.
Dean of FBFS

prof. Ing. Dušan Igaz, PhD.
Dean of FHLE

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SECTION

Biotechnology

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One pot synthesis of sialyllactose by magnetically immobilized inclusion bodies using cyclic regeneration of CTP

Martina BELKOVÁ, Romana KÖSZAGOVÁ, Jozef NAHÁLKA

Supervisor: Ing. Jozef Nahálka, PhD.

Human milk contains wide spectrum of sialylated olichosaccharides, which have significant benefits for infant´s health. They undoubtedly support development of nervous, gastrointestinal and immune systems, as well as resistance to pathogens. More than 70% of sialic acid residues are bond to oligosaccharides by sialyltransferases, whereas sialyllactose is the most abundant. Despite the natural presence of sialyllactose in human milk, its commercial production must overcome several hurdles. Key intermediate product in sialyllactose synthesis pathway is CMP-sialic acid, synthetized by fusion of CTP and sialic acid by CMP-sialic acid synthetase. Active inclusion bodies (IBs) are insoluble protein aggregates, previously considered as inactive waste in recombinant proteosynthesis. Since discovery of their partially remained activity, IBs can nowadays be used as biocatalysts in biochemical reactions. This new *in vitro* strategy can replace bacterial *in vivo* expression systems. One pot synthesis represents novel approach of combining several types of catalytic IBs in single reaction. Natural *in vivo* immobilization of IBs can be strengthened by additional immobilization by magnetics particles. This procedure allows reutilization of catalytic IBs and synthesis of product in several reaction cycles. For purpose of this experiment, mixture of iron-nickel magnetic particles was used to immobilize active IBs of polyphosphate kinase, cytidilate kinase, sialic acid aldolase and CMP-sialic acid synthetase. Only sialyltransferase was used in soluble form. To lower reaction costs, cyclic CTP regeneration from polyphosphate and CMP was used. Synthetized sialyllactose was observed by capillary electrophoresis and confirmed by TLC. To prevent accumulation of polyphosphate during several reaction rounds, and thus inhibiting overall efficiency of one pot synthesis, two strategies were compared. The starting concentration of polyphosphate in each sample was 2%. In first strategy, 12-hour break between reaction cycles was implemented, where immobilized IBs with remaining polyphosphate were placed in cold storage in TrisHCl buffer (100mM, pH 8). The second strategy used decreasing polyphosphate concentration by 0,5% from 2nd cycle, dropping to 0.5% at 4th and later cycles. In control sample, polyphosphate concentration remained stable at 2% each round. According to average yield 5.52 mM sialyllactose in five reaction cycles, the second strategy appears to be the most efficient. By rinsing with TrisHCl buffer and thus intermitting the reaction cycles, only 3.85 mM of sialyllactose was achieved, whereas, control sample with similar average yield 4.46 mM of sialyllactose in five reaction cycles was observed.

Key words*:* *active inclusion bodies, one pot synthesis, sialyllactose, immobilization, reutilization, CTP regeneration*

Acknowledgement: Work was supported by project APVV-18-0361

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Effect of sea buckthorn extract on the development of cataracts in ZDF rats

Jana HRNKOVÁ

Supervisor: prof. Ing Marcela Capcarová, DrSc.

Cataract, one of the leading causes of preventable blindness worldwide, results from lens degradation that leads to blurry vision. Because the majority of the population has limited access to cataract surgery, many plant extracts with anti-cataract compounds are tested. Sea buckthorn (*Hippophae rhamnoides*) belongs to one of the most valuable plants in the world. It contains more than 200 bioactive substances, all vitamins soluble in water and fats, bioflavonoids, amino acids, minerals and microelements. Curable effects of sea buckthorn can be used in many fields of medicine. The work focuses on the influence of sea buckthorn extract on the development of cataracts in the Zucker diabetic fatty (ZDF) (*fa/fa*) rats. 12-week-old males (n = 50) were divided into 6 groups, of which 2 diabetic controls were administered water and metformin (150 mg / kg body weight / day) and 4 experimental groups in which the animals were administered sea buckthorn by gastric tube in different concentrations, with or without added metformin (E1 = sea buckthorn 500 mg / kg body weight; E2 = sea buckthorn 500 mg + metformin 150 mg / kg / day; E3 = sea buckthorn 1000 mg; E4 = sea buckthorn 1000 mg + metformin 150 mg / kg / day). Cataract development was macroscopically monitored by magnifying glass along with photo documentation every 2 weeks for the duration of the experiment. The aim was to record the onset of cataract formation and the rate of progression to a significant stage. A scoring system was used for evaluation, which divided the cataracts into 5 stages, according to the degree of the lens opacification. The total number of cataract eyes and the representation of individual stages in the tested groups were evaluated. Interim results indicate that high levels of natural antioxidants in sea buckthorn could have the potential to delay cataract development. While the onset of cataracts due to hyperglycaemia was observed in diabetic animals in the control group, which was administered water, feeding of sea buckthorn delayed both onset and progression of cataracts. Further research will be necessary to understand the exact mechanism of the impact of individual antioxidants on pathological processes, as well as to determine the optimal dose of substances while maintaining safety for the organism.

Key words*: cataracts, lens, sea buckthorn, antioxidants, ZDF rats*

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Genetic diversity of common bean determined by SCoT markers

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Supervisor: prof. RNDr. Zdenka Gálová, CSc.

Common bean (*Phaseolus vulgaris* L.) is a legume of global importance. Bean is considered to be a valuable crop used for direct human consumption. Due to its optimal content of proteins and other essential compounds common bean has a potential as a functional food, especially in developing countries of Latin America and eastern Africa. Considering the growing global population and growing demands on the quantity and quality of food, many breeding programs focus on obtaining information valuable in breeding process of the economically important crops. The study of polymorphism and genetic diversity using DNA markers makes it possible to differentiate and to some extent characterize the genotypes of common beans. This information can be further used by breeders in order to improve the traits of existing bean varieties, and to obtain new bean varieties in the process of marker-assisted selection. The aim of our study was to analyze genetic diversity among 34 genotypes of common bean, from which 24 genotypes originated in 18 different countries and 10 genotypes were of unknown origin, based on DNA polymorphism using Start Codon Targeted markers (ScoT). Genomic DNA was extracted from leaves of 14-day old plants. A total of five SCoT primers (SCoT2, SCoT3, SCoT19, SCoT34, SCoT59) were selected for the present study. The number of DNA bands using individual SCoT markers ranged from 12 to 22. Overall, we observed 82 types of evaluable DNA bands, whereas 67% of bands were polymorphic and 33% of all bands were monomorphic. A dendrogram was constructed based on hierarchical cluster analysis using the unweighted pair group method with arithmetic average (UPGMA). The analyzed bean genotypes were divided into two main clusters, with variety Albena being separated in the second cluster from the remaining 33 varieties grouped in the first cluster further divided into subclusters. The dendrogram shows that the variety Michael of the unknown origin and variety Fruca Simpla from Italy are the most genetically similar. We also evaluated average diversity index (0.804), polymorphic information content (0.802) and probability of identity (0.013). According to the polymorphism detected by ScoT markers we can conclude that SCoT markers are useful tool for differentiating genotypes and assessing the genetic diversity of common bean genetic resources.

Key words*:* *bean, polymorphism, diversity, SCoT*

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Regulation of GIGANTEA genes in potato

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Supervisors: Zsófia Bánfalvi, Anikó Veres

GIGANTEA (*GI*) is a plant-specific nuclear protein, which was identified and isolated from several plant species from monocots to dicots. This gene is involved in various processes as for example, flowering time regulation, control of circadian rhythm, stress responses and, in potato (*Solanum tuberosum* L.), tuberization. The *GI* genes have daily cycle of expression. Although, no studies on the regulation of *GI* gene expression in potato has been published so far. The aim of our study was the analysis of the *GI* promoter sequence *in silico* and the expression of *GI* by quantitative reverse transcription PCR. We identified two *GI* genes homologous to *Arabidopsis* in potato on chromosome 4 and 12 (*StGI.04* and *StGI.12*). The *GI* promoter regions of the commercial potato cultivar 'Désirée" were cloned and found to be almost identical with the genome-sequenced *Solanum phureja GI* promoter sequence. We found more than ten transcription factor families binding to the *GI* promoters. In both *GI* promoters, an EVENING ELEMENT (EE) and ABSCISIC ACID (ABA) RESPONSE ELEMENT LIKE elements connected to circadian regulation and a binding site for POTH20 probably involved in tuber initiation were detected. Nonetheless, location of these elements and several *cis*-acting regulatory elements in the promoter regions of the two *GI* genes were different. The osmotic stress as well as heat and cold up-regulated *StGI.04*, while the same stresses down-regulated the expression of *StGI.12*. ABA had no effect on *StGI.04*, but induced *StGI.12*. All these results indicate that *StGI.04* and *StGI.12* may have different functions. This research lays foundation for further examination of the roles of *GI* genes in potato.

Key words*: Solanum tuberosum GIGANTEA, promoter elements, transcription factors, circadian rhythm*

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Genetic variability of *Fagopyrum* genotypes determined by gene specific markers

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Supervisor: doc. Mgr. Želmíra Balážová, PhD.

Fagopyrum represents economically and nutritionally important crop which belongs to the group of pseudocereals. Buckwheat is referred to be a functional plant with variety of uses in the world. Grains of *Fagopyrum* are nutritionally significant. They are characterised by a high content of starch, proteins, flavonoids and fibre. *Fagopyrum tataricum* is characterised by a bitter taste but also has a higher total content of B vitamins and higher level of antioxidants, especially rutin in comparison to *Fagopyrum esculentum*. Genetic diversity refers the amount of genetic variability between genotypes or species. Molecular methods are important part of the most of genetic studies concerning to distribution of genetic diversity and breeding analyses. Amplifications techniques based on the polymerase chain reaction are important for molecular research. A gene specific SCoT technique was applied for studying of the genotypes of *Fagopyrum*. SCoT technique is used for the detection of polymorphism in plant genes and for evaluation of genetic diversity and mapping the functional regions of the genome. The aim of the work was to perform molecular analyses of 21 selected genotypes of *Fagopyrum esculentum* and 14 selected genotypes of *Fagopyrum tataricum* by using four gene-specific SCoT markers. The seedlings of buckwheat were used for DNA isolation. Isolated DNA was subsequently used to amplify DNA fragments by using a polymerase chain reaction. Total number of fragments was 79 of which 75 fragments were polymorphic. The average number of polymorphic fragments was 18.8. SCoT 12 and SCoT 29 were considered as the most polymorphic markers for which percentage of polymorphism was 100%. The lowest percentage of polymorphism was shown by SCoT 26 (81.3%). Polymorphic information content (PIC) characterized polymorphism of used SCoT markers. The PIC value ranged from 0.887 (ScoT 26) to 0.929 (ScoT 29). Genetic diversity of buckwheat was determined by using hierarchical cluster analysis using UPGMA algorithm in the created dendrogram. Genotypes of buckwheat divided into two main clusters in the dendrogram. Two genotypes of *Fagopyrum esculentum* (Siva, Špačinska I) from Slovenia and the Slovak Republic, respectively, as well as another two *Fagopyrum esculentum* genotypes (Bogatyr and Darina) from Russia and Slovenia, respectively, were genetically the closest. According to our results, we can consider SCoT technique as appropriate for differentiation of genotypes of *Fagopyrum esculentum* and *Fagopyrum tataricum*, which demonstrate high average percentage of polymorphism (95%) and high average PIC value (0.908).

Key words: *Fagopyrum esculentum, Fagoprum tataricum, polymorphism, SCoT markers, dendrogram*

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The promoter analysis and gene expression of potato BIG BROTHER gene

Khongorzul ODGEREL

Supervisors: Zsófia Bánfalvi, Anikó Veres

The RING finger protein *BIG BROTHER/ENHANCER OF DA1 (BB)* has ubiquitin E3 ligase activity and is a central growth regulator in *Arabidopsis*. *BB* restricts cell proliferation and promotes leaf senescence. Homozygous *BB* mutant (*bb-1*) formed larger petals and sepals, as well as thicker stems than wild type. Some RING finger proteins are strong candidates for being negative regulators of stress responses. However, no such studies related to *BB* gene have been reported thus far. To our knowledge the *BB* gene of potato (*Solanum tuberosum* L.) has not been studied either. Thus, our research work was aimed to analyze the *BB* promoter sequence *in silico* and study the *BB* expression in different organs and under different stress conditions by quantitative reverse transcription PCR (qRT-PCR) in potato. We identified a *BB* gene homologous to *Arabidopsis BB* in the genome-sequenced *Solanum phureja* on chromosome 11 (*StBB*) and predicted transcription factor (TF) families binding to the *BB* promoter including the TFs RELATIVE OF EARLY FLOWERING 6 (*REF6*), ABSCISIC ACID INSENSITIVE 3/VIVIPAROUS 1 (ABI3/VP1) and MIKC-type MADS (MIKC_MADS) family proteins involved in leaf, lateral root, carpel and stamen development and maintenance of floral organ identity. Furthermore, binding sites for a basic helix-loop-helix (bHLH) TF connected to the cold response and circadian rhythm and a myeloblastosis (MYB) family protein involved in salt stress were detected. The qRT-PCR results showed that *StBB* expression was up-regulated by the salt (NaCl) and osmotic (PEG) stresses in leaves. However, abscisic acid (ABA), cold (4 °C,) and heat (42 °C) treatment did not show any effect on *StBB* expression. The *StBB* gene was expressed in all tested organs. The highest expression was detected in petal and stamen, while roots and stems expressed lower level of *StBB* than the floral organs.

Key words*: Solanum tuberosum* L., *BIG BROTHER gene, growth regulator, promoter elements, transcription factors, abiotic stress*

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Application of green sulphur bacteria *Chlorobium limicola* for detoxifying of H_2S in wastewater and the perspective of subsequent use for glycogen production

Martin STRUK, Ivan KUSHKEVYCH

Supervisor: doc. Ivan Kushkevych, Ph.D.

In rural areas, the uncontrolled discharge of agricultural and agro-industrial wastewaters is causing the contamination of water bodies with pathogens and toxic pollutants, such as hydrogen sulphide. The most common treatments of H_2S are physicochemical processes. However, most of these technologies are unsuitable for small-scale applications. The alternative to these processes are biological treatment methods such as use of bacteria. The green sulphur-oxidizing bacteria (GSB), mainly genus *Chlorobium,* are capable of H_2S removal. In the process of anoxygenic photosynthesis, the hydrogen sulphide is oxidised and accumulated as elemental sulphur outside of the cell. The aim of this ongoing research is to design and test photobioreactor concepts and evaluate the ability of *Chlorobium limicola* DSM258 to remove H_2S from wastewater under various conditions as well as asses the possibility of glycogen production. Results from our previous experiments with synthetic biogas are showing the successful application for H_2S removal. The 2 L fed-batch/semi-continuous reactor was able to remove 100% of H_2S from gas stream (S^{2-} intake of 0.024 $nM \cdot d^{-1} \cdot L^{-1}$). However, adapting the process for wastewater treatment requires changes in process design. The first proposed configuration is a bioreactor consisting of long Tygon tube wrapped around construction and the necessary light is supplied via LED or incandescent lamp. The second configuration uses cylindrical photobioreactor with a magnetic stirrer at the bottom. The light is provided by optical fibres inside the reactor. They also provide a surface for biofilm formation. The first test runs were performed with cylindrical photobioreactor under external illumination (LED bulb; 4000–5000 lux) in batch mode. Operating conditions were monitored and kept at pH = 6.6–7.0; ambient temperature (23–25 °C) and stirring 250–300 rpm. Oxygen content in reactor was controlled by sensor and by manually flushing the reactor with N_2 when necessary. Bacterial growth was measured as an absorbance of biomass at 650 nm. In the first two days, the initial content of H_2S in medium decreased by 85% in first test run and by 100% in the second. The H_2S was replenished on 5^{th} and 6^{th} day in first experiment and reached 0% content on 1^{st} and 2^{nd} day respectively after its injection. Absorbance of the bacterial culture on the second day also decreased for both experiments. For the rest of the experiments, only negligible rise of absorbance was measured, and no visible growth was observed. High concentration of oxygen (> 10%) was detected in the Run 1 after 28 h and after 72 h in run 2. The probable causes of growth inhibition were the unsuitable light (commercial LED bulb) and minimal mineral medium which in combination were not able to provide suitable growth conditions. Presence of oxygen in reactor on 2^{nd} and 3^{rd} day could be another reason and can explain the sharp decrease of H_2S content despite no apparent growth of bacteria. Possible causes of its presence, such as airtightness of reactor vessel or leaks in tubes supplying the N_2, are under investigation. The results gathered up to now are indicating that further testing and adjustment of parameters are required for evaluation of effectiveness of these designs.

Key words: *hydrogen sulphide removal; phototrophic sulphur bacteria, bioreactor, wastewater purifying*

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Antifungal activity of Green Mandarin essential oil against selected *Penicillium* species causing spoilage of bakery products

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Supervisor: prof. Ing. Miroslava Kačániová, PhD.

The current study was aimed to assess antifungal activity of Green Mandarin (*Citrus reticulata*) essential oil (GMEO) acting as the vapor phase in different concentrations (62.5, 125, 250, and 500 mL/L) against two selected *Penicillium* (*P.*) species (*P. expansum* and *P. chrysogenum*) growing on wheat bread. The volatile compounds of GMEO were identified by Gas Chromatography-Mass Spectrometry (GC-MS). Moreover, the antioxidative properties of the GMEO were determined using DPPH radical scavenging assay, and its *in vitro* antifungal activity was evaluated using the disc diffusion method. Bread loaves were baked in experimental conditions, and *in situ* antifungal activity of GMEO was estimated using stereological approach. Our data showed that the major constituents of *Citrus reticulata* EO were α-limonene (71.5%), γ-terpinene (13.9%) and β-pinene (3.5%). The GMEO exhibited weak antioxidant activity with the value of inhibition 5.6 ± 0.7%, which corresponds to 103.0 ± 6.4 μg TEAC / mL. Evaluation of GMEO *in vitro* antifungal activity revealed that inhibition zones ranged from 0.00 ± 0.00 (no antifungal effectiveness) to 5.67 ± 0.58 mm (moderate antifungal activity). From *in situ* analysis, it was evident that 250 μL/L of GMEO displayed the lowest value for mycelial growth inhibition (MGI) of *P. expansum* (-51.37 ± 3.01%) whose negative value even indicates a supportive effect of the GMEO on the fungi growth. On the other hand, this concentration (250 μL/L) of GMEO had the most inhibitory impact (MGI: 54.15 ± 1.15%) on the growth of *P. chrysogenum*. In this context, our results allow for the conclusion that the application of GMEO in the vapor phase seems to be an effective only against *P. chrysogenum* suggesting GMEO to be a sustainable alternative to the use of chemical inhibitors for bread preservation.

Key words*: Citrus reticulata, volatile compounds, antioxidant activity, disc diffusion method, antifungal properties, bread*

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Effect of the equilibration time on post-thaw quality of manually freezed ram sperm

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The aim of our research was to improve ram sperm post-thaw quality through optimization of cryopreservation regimen. We tested different lengths of equilibration (EL; 0, 2, 4, 6 and 8 hours) of ram sperm cryopreserved using manual slow freezing technique in the liquid nitrogen (LN2) vapours. The ejaculates were collected from clinically healthy and sexually mature rams (n = 12) of Wallachian sheep breed (2.5-5 years) twice a week by an electro-ejaculation within the autumn season. Sperm doses were extended in a Triladyl® diluent containing glycerol, egg yolk and antibiotics. Following thawing, the sperm samples were analysed for the motility (CASA), viability (SYBR-14/PI) and *in vitro* penetration/fertilization (P/F) assays. It was found that the EL of 6 hours (EL-6) ensured higher post-thaw sperm total (76.6±2.3%) and progressive (65.0±4.18%) motility compared to other lengths tested. Using a SYBR-14/PI assay, the proportion of viable versus dead spermatozoa (47.67% vs. 31.11%, resp.) in the cryopreserved group significantly differed from those in the control group (79.86% and 14.07%, resp.). Fertilizing ability of cryopreserved sperm (EL-6), examined by *in vitro* heterologous P/F test on bovine mature oocytes (62.00%), was significantly lower compared to the control group (72.82%). However, in the percentage of fertilized zygotes the EL-6 group (54.67%) did not differ from the control (52.26%). These results provide evidence that extension of equilibration time up to 6 h can improve post-thaw quality of ram sperm.

Key words*: sheep, sperm, manual freezing, equilibration, motility, viability*

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SECTION

Animal Production

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Influence of sex on meat performance of geese with semi-intensive fattening

Marie HAMADOVÁ

Supervisor: doc. Ing. Henrieta Arpášová, PhD.

The aim of the study was to compare the carcass characteristics in male and female of Slovak goose in half-intensity fattening at climatic conditions of south-western Slovakia. During period to 12 weeks, the 40 one-day-old goslings were kept in littered pens, the floor space was 6 birds per m^2. From 13 to 16 weeks, goose had access to the pasture during the day, and were closed in shelters at night. Goose had *ad libitum* access to water and feed: complete commercial diets (0–12 weeks), pasture with additional feeding was performed with corn (200 $g.day^{-1}$) in the mornings and evenings (13–16 weeks). On 112^{th} day, 5 males and 5 females were chosen for slaughter. The abdominal fat, giblets, breast muscle were weighed separately. Thighs were deboned to obtain the weights of thigh muscle. The carcass yield was calculated as the percentage of cold carcass weight from slaughter weight. The percentages of breast and thigh muscle, abdominal fat and giblets were calculated as a percentage of the cold carcass weight. Significant differences ($P<0.05$) were observed between males and females in slaughter weight (5343.89 g vs. 4881.26 g) and carcass weight (4052.67 g vs. 3665.14 g). The sex of goose had no significant effect ($P>0.05$) on the muscles from breast (17.48% vs. 17.52%) and thighs (11.19% vs. 11.22%), abdominal fat (3.07% vs. 3.11%) and carcass yield (70.89% vs. 71.03%). The proportion of liver in the carcasses of males and females were similar ($P>0.05$; 5.32% vs. 5.28%). Gizzard percentage was significantly higher ($P<0.05$) in males (4.28% vs. 4.05%), and heart percentage was significantly higher ($P<0.05$) in females (0.81% vs. 0.89%).

Key words: *goose, sex, carcass, breast muscle, thigh muscle, giblets*

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Evaluation of behavior and physiological traits in calves during two hours after birth

Terézia HEGEROVÁ

Supervisor: doc. Mgr. Peter Juhás, PhD.

In the present experiment, we focused on the evaluation of the behavior of Holstein calves in the first two hours after birth. The research took place at the Slovak University of Agriculture Farm of in Oponice. Behavior was recorded by two IP cameras placed above calving pen. The calf sample consisted of 11 individuals. Behavior records were analysed by BORIS software. Behavior was evaluated by next traits: total duration of lying, standing and moving, average duration of one bout lying, standing and moving, percentage rate of lying, standing and moving, number of occurrence of bouts for all scored traits, mean duration inter-event interval for all scored traits. The basic physiological parameters (heart rate -HR and rectal temperature –RT) were measured in four 15 minute intervals during first hour after the birth (M15, M30, M45 and M60). Difference in behavior between first and second hour of observation was tested by non-parametric Kruskal Wallis test. Relation among measured and scored traits was analysed by Spearman rank correlation. The average length of birth was 1 hour and 4 minutes (s.d. = 0.33 seconds). The first activity after birth in the calves were attempts to lift up head. The calves in the test sample had a latency to stand up 11 minutes and 57 seconds (s.d. = 22 minutes 53 seconds). The rectal temperature of the calf after birth decreases with time, the highest values were measured at after 15 minutes M15 (39.31 °C, s.d. = 0.62 °C), lowest at M45 (38.68 °C, s.d. = 0.50 °C). Unlike temperature, heart rate does not decrease over time. The highest average heart rate values were measured at M30 (163.27 beats per minute (BPM), s.d. = 13 BPM) and the lowest at M15 (160.36 BPM, s.d. = 12.45 BPM). When comparing activity between the first and second hour, we found that calves are more active during the second hour after birth. In the first hour the calves lie longer ($\bar{x}$ = 3351.18 seconds, s.d. = 400.62 seconds) than in second hour ($\bar{x}$ = 2481.70 seconds, s.d. = 1067.22 seconds). Difference in lying, standing and moving between 1st and 2nd hour after birth is significant ($P < 0.05$). Positive correlations were found between the heart rate and the number of standing periods, calves with a higher number of standing periods ($r = 0.691$ $P < 0.05$) and the period of movement ($r = 0.647$ $P < 0.05$) had a higher heart rate. We can conclude there is relation between signs of activity in calves in first hour after birth as well as relation between activity and hear rate.

Key words: calves, Holstein, physiological indicators, behavior

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Analysis of selection signatures in sheep using variation in linkage disequilibrium

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Supervisor: prof. Ing. Radovan Kasarda, PhD.

The aim of the work was to identify genomic regions undergoing selection, and therefore increase our knowledge about the Valachian sheep. The Valachian sheep is a Slovak national, indigenous sheep breed. It is an endangered, but highly important animal genetic resource, therefore its genomic characterization is of upmost importance. A total of 97 Original Valachian and 69 Improved Valachian sheep were genotyped using the GeneSeek GGP Ovine 50K chip. The data was quality controlled using PLINK, and animals with more than 10% missing SNPs, as well as individual SNPs with more than 10% missingness, those below 1% minor allele frequency and not following the expected Hardy-Weinberg equilibrium distributions at $p < 10^{-7}$ were excluded from the data set. The SNPs from unplaced or sex chromosomes were also removed. After quality control there were 96 animals and 38236 SNPs remained for the Original Valachian sheep, and 68 animals and 39240 SNPs were left for the Improved Valachian sheep population. From these 37788 SNPs were present in both data sets, which was used to detect selection signatures. The VarLD software was used to compare linkage disequilibrium patterns in the two populations, which were then further analyzed and visualized using the R programming language. The highest signal, with the top 0.01% of the results was found between 10.8 and 11.0 Mb on chromosome 22. This relatively narrow region contained five protein coding genes and three pseudogenes. The LIPA gene was found to be involved in the lipid mechanism and composition of lipoproteins, response to wounds and inflammations, but also in molecular genetic mechanisms affecting fecundity in sheep. The family of IFIT genes in the region contribute to parasite resistance, while the SLC16A12 gene to resistance to bacterial infections. It can be concluded that all genes in this highly selected region point towards traits related to immunity, which confirms the adaptation mechanisms of the Valachian sheep to the harsh environmental conditions.

Key words*: genomics, selection, single nucleotide polymorphism, sheep*

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Heterozygosity rich regions in Slovak Spotted cattle

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Supervisor: prof. Ing. Radovan Kasarda, PhD.

Heterozygosity is defined as the possession of two different alleles of a particular gene or genes by an individual. Heterozygosity the same as homozygosity is related to inbreeding and genetic diversity. The aim of the study was to analyse heterozygosity rich regions (HRR), which are regions with high rates of heterozygosity. HRR in genotyped Slovak Spotted cattle was studied. 87 animals were genotyped by BovineSNP50 BeadChip. We have removed 2 individuals due to missing genotype data. There were 85 animals (37 bulls, 48 cows) with 38498 SNP in final dataset. For ROHet analyse following set of parameters was used: 15-SNP sliding windows, minimum number of heterozygous SNP in the window was 15, maximum number of SNP with opposite genotype 3, maximum number of missing genotypes 2, maximum gap between consecutive SNPs 1Mb and minimum length of the run 250 kb. In our population of Slovak Spotted cattle were detected 4142 HRR. 4055 HRR were in 0-2 Mb class (1644 at bulls, 2411 at cows) and only 87 HRR in 2.4 Mb class. The most HRR were on chromosome 1 (323 – 15.5%) and least was on chromosome 27 (81 – 2.6%). The largest region was located on chromosome 21 (1956.692 kb) and the shortest on chromosome 7 (53.778 kb). Chromosomes 2, 4, 7, 14, 16, 17, 20, 23 had 1 region, chromosomes 3, 9, 10, 21 had 2 regions and chromosomes 11 and 1 had 3 regions. In HRR we found 62 genes with known and unknown function. There were some regions with no genes recorded. Authors of another studies hypothesized, that ROHet could be linked with balancing selection. ROHet can include loci, that contribute to important functions (fertility, fitness, survival rate). In these segments of the genome with important functions can be diversity beneficial. Results of our study will help to make breeding strategies to maintain and improve genetic diversity of Slovak Spotted cattle.

Key words*: Slovak Spotted cattle, heterozygosity rich regions, biodiversity*

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Mineral status of young horses in a selected area

Dušan SOLÁR

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A balanced and sufficient diet plays an important role in the proper development of young horses. The suitable location for grazing, especially the soil composition could be crucial for nutritional value of pasture. The aim of the study was to analyse a mineral profile of horse blood serum in relation to selected area. Blood serum of eight males (geldings and stallions) and eight mares grazed in selected area in Nové Mesto nad Váhom district were collected. The biochemical analysis of horse blood serum for determination of mineral concentration was conducted using Rx Monza Clinical Chemistry Analyser (RANDOX Laboratories, Ltd.) Level of sodium (Na), potassium (K) and chlorides were determined using an automatic analyser EasyLytePlus (The Hague, Netherlands). We found that the average level of calcium (Ca) in horse blood serum was 3.02 mmol/L, which is within the reference range. The level of phosphorus (P) was also within the reference interval whereas the values were in a range of 0.63 to 1.05 mmol/L. The high imbalance occur in the level of magnesium (Mg), when the level in blood serum of mares (1.19 mmol/L) was significant higher than reference range. The Na level in horse blood serum was in range of 135.9 mmol/L to 145.5 mmol/L and it reached the upper limit of reference range. Conversely, we detected a low level of K at the lower limit of reference range. The differences in mineral parameters in blood serum of observed horses were statistically significant. According to analysed values of chlorine (Cl), which level was in the range of 99.0 mmol/L to 110.41 mmol/L, we can conclude that the examined horses reached higher proportion of this indicator with a slight exceeding of the reference range. The differences in Cl levels in blood serum of horses were not statistically significant. According to analysis of mineral profile of horse blood serum it may be concluded that the mineral status of tested horses in selected area is unbalanced. It could have a strong negative effect on further development of young horses.

Key words: young horse, blood serum, mineral profile

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Smart hive

Marek URBAN

Supervisor: doc. Ing. Róbert Chlebo, PhD.

Beekeeping same as any other industry keeps modernizing using technologies that help us to acquire more honey, pollen, healthier and happier bees etc. Latest inventions can be used to track the conditions of bee colonies and beekeeper can decide whether or not to do certain interventions that are needed to improve the comfort of bees. Modern technologies are becoming cheaper and more accessible to everyone. They're easier to use and to understand. Smart hive can show the beekeeper when the queen starts laying eggs after winter or the death of the colony, it could predict the swarming, can detect nectar flow start/end, starvation, broodless colony or drone-laying colony, pollen collection, varroa infestation or illnesses. All these information can be provided at distance, without manual inspection which is even more important for beekeepers that travel with their bees to distant apiaries to obtain more honey during the blooming season. It can be used for scientific purpose so we can understand bees better or simply by people that love nature and bees and want to observe the way how they live. With help of our smart hive with temperature and humidity sensors inside as well as outside of the hive and a scale measuring its weight we were able to check the status of the bee colony during the whole year. Results showed that the temperatures outside during season varied from 7 °C up to 42 °C and the humidity from 30% up to 100% during rain the temperature inside the colony with brood was always 33–36 °C and the humidity around 65%. During winter the temperature inside the hive, close to cluster was 15–20 °C even when outside temperature often dropped down to -10 °C. When the colony didn't survive the harsh winter we could immediately see that the temperatures and humidity inside and outside of the hive were almost the same. Also when colony swarmed the sudden huge drop of weight on the scale in short time alarmed us on what happened and we could intervene. Adding more sensors measuring more parameters could help us collect more data and in the future even predict bee colony problems such as swarming, varroa infestation, hunger and many more.

Keywords: precision beekeeping, smart hive, IoT, sensors

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SECTION

Applied and molecular biology

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Isolation of trypanosomatids respiratory chain enzymes

Katarína BEHINSKÁ

Supervisor of project: RNDr. Petra Čermáková, PhD
Actual supervisor: prof. Ing. Adriana Kolesárová, PhD.

Complexes I and II are the gateway of electrons into the respiratory chain. Complex I transfers electrons from NADH to ubiquinone and pumps the protons across the inner mitochondrial membrane. Complex II receives electrons from succinate and transfers them to ubiquinone without forming a proton gradient. In trypanosomatids, these enzymes are much larger than in other eukaryotes – in the case of complex I, genomic analyses showed more than 60 subunits and complex II is composed of up to 12 subunits. In species that do not have complex I in their respiratory chain, it is thought that complex II transfers electrons to ubiquinone. The plant parasite *Phytomonas serpens* has the best studied trypanosomatids complex I. Proteomic analysis of its multi-subunit enzyme purified by native gel electrophoresis confirmed the presence of 32 subunits. In this study we tried to isolate this enzyme by ion exchange chromatography. We obtained a purified fraction with a degree of purification greater than 5 and we were able to confirm the presence of another 5 subunits of this complex including one subunit encoded by mitochondrial DNA (NDUFS6, NDUFB9, NDUTB11, NDUTB30 and ND7). In *Kentomonas sorsogonicus* and *Wallacemonas raviniae* (do not have complex I) we measured several times higher activity of the complex II than in *Sergeia podlipaevi* and *Novymonas esmeraldas*, in which the first enzyme of the respiratory chain is active. We have also shown that the 440 kDa protein ladder of *K. sorsogonicus* is actually the complex II. This is the first resolution of this enzyme complex on the profile of the second dimension.

Key words: *complex I, NADH dehydrogenase, complex II, succinate dehydrogenase, Phytomonas serpens, Kentomonas sorsogonicus*

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Differences in the oxidative profile between *in vitro* capacitated and cryocapacitated bovine spermatozoa

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Supervisor: prof. Ing. Norbert Lukáč, PhD.

The success of cryopreservation depends mainly on the cell sensitivity to low temperatures during freezing and their viability after thawing. The cryopreservation process may cause plasma membrane damage, reduction of motility, induction of premature capacitation and increase the generation of reactive oxygen species (ROS). The aim of our study was to compare the oxidative profile between *in vitro* capacitated and cryocapacitated bovine spermatozoa. Cryo-capacitation is one of the major factors associated with reduced fertility of frozen-thawed semen used for artificial insemination. For our experiment we used semen samples from adult Holstein bulls (n=30) obtained from a local farm. All samples were divided into three parts, the first part was incubated in physiological saline solution as control (CTRL) while the second part was incubated in a capacitation medium (CAP) for 30 min. at 39 °C and 5% CO_2 concentration. The last part of each sample was cryopreserved (CRYO) and stored in liquid nitrogen at -196 °C for further analysis. The quantification of ROS, hydrogen peroxide a hydroxyl radical generation was measured by using combined spectro-fluoro-luminometer Glomax Multi+. The analysis of superoxide production was performed with the NBT-test and optical density was evaluated spectrophotometrically with the Glomax plate reader. Based on the results, the CRYO group showed a significant increase ($P<0.0001$) in the concentrations of ROS, hydrogen peroxide and hydroxyl radical against CAP or CTRL group. However, the amount of superoxide radical was statistically higher ($P<0.0001$; $P<0.001$) in the CAP group when compared to CRYO and CTRL. In conclusion, cryopreservation leads into cryocapacitation, which negatively affected almost all parameters in the oxidative profile of bovine spermatozoa under *in vitro* conditions and increase the risk of oxidative stress development.

Key words: *bull, spermatozoa, cryocapacitation, cryopreservation*

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Prediction of miRNAs with target sequences involved in the cyanogenic glycosides and lignans metabolism by in silico approach

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Supervisor: prof. Ing. Katarína Ražná, PhD.

Molecules micro RNAs (miRNAs) are endogenic 19-24 bp long noncoding single strands. Their main role lies in negative regulation of gene expression by base-pairing with target post-transcripted mRNAs and supporting of their cleavage. MiRNAs occur within whole genome and current bioinformatic methods enable to search them in open-source sequential data. Based on their high conservative features it is possible to compare them with another plant species and/or find miRNAs according to target gene sequence. The aim of research was to predict specific miRNAs which are complement to genes involved in metabolism of plant cyanogenic glycosides and lignans. *Linum usitatissimum* L. was chosen as a plant of interest but there were also searched many other plant genes and miRNAs depending on available databases. The sequences of 6 target proteins were found on NCBI database (https://www.ncbi.nlm.nih.gov/) and searched in the flax genome with use of the Plant Comparative Genomics portal Phytozome 13 (https://phytozome-next.jgi.doe.gov/). All sequences of 48 885 mature miRNAs and 38 589 hairpin structures were downloaded from online database miRbase (www.mirbase.org). The 9 678 hairpins structures assigned with 11 916 mature miRNAs were identified in flax genome. They were subsequently aligned with target genes by several different algorithms such as psRNATarget (https://www.zhaolab.org/psRNATarget/), Single sequence searches of miRbase database and BLASTn tool (https://blast.ncbi.nlm.nih.gov/). The results of potential miRNAs were compared to each other on the base of their alignment score. The outcome of this research was prediction of new potential miRNAs and their target genes in flax genome with aiming to lignan´s and cyanogenic glycosides´ metabolic pathway.

Key words: *miRNAs, in silico, target gene, cyanogenic glycosides, lignans, flax*

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The effect of polyphenolic biomolecules on sperm motility

Michal LENICKÝ

Supervisor: Ing. Eva Tvrdá, PhD

The surrounding world is very diverse and offers a wide range of bioactive substances with a direct impact on human health. Recent research indicates that the group of polyphenolic substances contains a wide range of beneficial effects such as antioxidant protection of cells from damage or anticancer protection. As male fertility defects are an increasingly actual topic, we have been interested in how polyphenols could affect sperm motility as one of the basic parameters determining the success of fertilization. In our study, we focused on a group of polyphenolic substances consisting of Epicatechin, Gallic Acid, Quercetin, Rutin, Kaempferol and Resvertrol obtained by lyophilization of grape peels. The study involved 48 men with an average age of 28.21 ± 6.65 years. Probands used polyphenolic supplements for 12 weeks, the exact daily dose was chosen individually based on the weight of the proband. Probands provided ejaculate samples obtained by masturbation at time 0 (control I.), 3, 6 and 12 weeks of consumption of a dietary supplement containing polyphenols, and after the next 12 weeks another sample was performed, which represented control no. II. Our aim was to find out how polyphenolic supplements affect sperm motility. This was determined using the CASA system (computer-assisted sperm analysis). After cell separation, we analyzed the zinc content from the seminal plasma. The concentration of zinc is extremely important for sperm and we were interested in how polyphenolic supplements will affect its concentration due to its irreplaceable role as well as the possibility of cytotoxicity. Its exact concentration was determined using a commercially purchased Dialab Zinc 5-Br-PAPS colorimetric assay. As sperm movement is a very energy-intensive activity, we also determined the concentration of Fructate and Citric Acid as very important substances of energy metabolism. Both were based on the colorimetric test Fructate assay kit and Citric acid assay kit were commercially purchased from Megazyme. The resulting absorbance was measured using an ELISA reader. After statistical data processing, we recorded a significant increase in Zinc concentration ($P < 0.05$) compared to control I. in all three samples (3,6,12 T.) during 12 weeks of use. We recorded a slight but insignificant increase in concentration compared to the control for citric acid ($P > 0.05$). Fructose concentrations ($P > 0.05$) remained almost unchanged at all sampling intervals. We also observed statistically insignificant differences in the case of sperm motility ($P > 0.05$), but we see that with increasing time of use, motility gradually increases in small pieces. In conclusion, it follows that more significant differences could be observed with longer use of polyphenolic supplements or in patients with reproductive dysfunction.

Key words*: polyphenols, seminal plasma, sperm motlitiy, zinc, fructate, citric acid*

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Comparison of effect of sea buckthorn extract and quercetin on human ovarian granulosa and cancer cells *in vitro*

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Sea buckthorn (*Hippophae rhamnoides* L.) is a good source of bioactive polyphenols for human consumption. The health benefits of sea buckthorn are due to the content of phytochemicals, mainly flavonoids and phenolic acids such as quercetin, rutin, isorhamnetin, catechin, and gallic acid, which are commonly presented in sea buckthorn extract. Recent studies have demonstrated the ability of sea buckthorn and its flavonoids to regulate the expression of anti-apoptotic and pro-apoptotic proteins in various types of cancer. It is well known that quercetin can decrease cell survival and viability of cancer cells. The aim of this *in vitro* study was to compare the effect of ethanol sea buckthorn extract at concentrations 0; 12.5; 25; 50; 100; 200 µg/mL (24 h) and quercetin at concentrations 5; 10; 20; 50; 100 µmol/mL (24 h) on the viability of human ovarian granulosa cell line (HGL5) and human ovarian carcinoma cell line (OVCAR-3), as well as secretion of steroid hormones (progesterone and 17ß-estradiol) by HGL5. Cell viability was evaluated by *AlamarBlue*^{TM} assay, and the release of steroid hormones was assayed by ELISA method. In our research sea buckthorn extract and quercetin did not significantly affect the viability of ovarian granulosa cells HGL5. However, quercetin at the concentration 100 µmol/mL significantly decrease ($P \leq 0.001$) viability of ovarian carcinoma cells OVCAR-3. Progesterone release by HGL5 cells was not significantly affected after sea buckthorn extract and quercetin addition. However, a significant ($P \leq 0.05$) decrease of the 17ß-estradiol secretion by HGL5 cells after sea buckthorn extract addition at the concentration 25 µg/mL was observed. On the other hand, a significant ($P \leq 0.05$) increase of the 17ß-estradiol secretion by HGL5 cells after quercetin (50 µmol/mL) addition was observed. Our results have shown that quercetin affected the viability of human ovarian carcinoma cell line and also secretion of 17ß-estradiol in human ovarian granulosa cell line. Sea buckthorn extract had an effect only on the secretion of 17ß-estradiol by human ovarian granulosa cell line while the viability of ovarian cells remained unchanged. Our findings suggest that sea buckthorn and quercetin may regulate processes of viability, steroidogenesis and apoptosis in dose-dependent and cell-specific manners, however further study is needed to clearly identify its bioactivity.

Key words: *sea buckthorn, quercetin, ovarian cells, cancer, steroid hormones*

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Chiral knots display symmetry breaking in helical confinement

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Topological knots naturally occur on DNA due to conformational moves or activity of molecular machines. Around half of knots are chiral including the simplest and most abundant trefoil knot. In some bacteria, occurrence of chiral knots is preferred over the achiral ones and in some one form of chiral knots is more abundant. Experimentally, it was successful to separate chiral knots through 2D-gel electrophoresis, where their structure was altered by negative supercoiling. In general, we know that chirality plays an important role in biological systems, yet there are still mysteries to uncover, especially regarding interaction of chiral molecules towards various environments. In this work, we observe differences between right- and left-handed forms of trefoil knots and their interplay with various confinements (cylinder, toroid, helix) through molecular dynamics simulations in ESPResSo. Simulations in cylinder provided general information about behaviour of knots in confined system, their size and shape properties, which we divided into two categories – geometrical properties independent of chirality and topological parameters influenced by handedness of the knot. Toroid confinement represents a system curved in space in one dimension. Such curving, however, was not enough to enhance distinguishability of chiral knots and they had similar topological properties as in cylinder. Right-handed helix, on the other hand, strongly influenced knot topology and induced symmetry breaking for chiral knots. Right-handed knot was delocalized and compressed at its centre, while the left-handed knot expanded its main loop towards confinement wall, making it more localized, spherical and better fitting in the helix. Different topological behaviour of chiral knots as well as introduced asymmetry visible on radius of curvature indicates that helical confinement is suitable for distinguishing and direct separation of right- and left-handed forms of knots.

Key words: *polymer, knot, chirality, confinement, DNA, MD simulations*

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Entropic competition between supercoiled and torsionally relaxed fibers drives chromatin loop extrusion

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In 1977 Paulson and Laemmli dissolved meiotic chromosome – the X shaped one – and discovered that it consisted of about 100 kilobase DNA loops attached to a protein scaffold. Immediately it came to the question, if these loops have a biological function. If such a biological function should exist, the loops would need to exist also in interphase chromosomes, i.e. during the longest period of the cell cycle when the chromosomes exist in form of globules and perform their basic biological functions. Experimental discovery of the loops in the interphase chromosomes was challenging and became only possible after developing a group of experimental methods for chromosome conformation capture – also named 3C. After improvement of the method's resolution in the new generation called Hi-C, the loops were independently discovered by three teams of authors in 2012. Next question that naturally occurred was how these loops are created. In our paper, we propose a novel Brownian ratchet mechanism for loop extrusion. Our model consists of fibre whose portion is stressed by axial rotations and a torsionally relaxed part. These two parts are separated by position of the "structural maintenance of chromosomes" (SMC) sliplink. The axial rotations mimic action of RNA-polymerase that performs transcription. The SMC is thermodynamically coupled with the fibre by exerting friction to axial rotations, thus preventing the supercoiling to escape. The supercoiled fibre can relieve from the increased energy only when the SMC moves further ahead from the transcription site and new portions of relaxed fibre flow into the emerging loop. The decrease in energy is temporary and soon replenished by the ongoing transcription and the SMC has to move again. Moreover, the entropic character of this process enables the loop extrusion to take place even if the SMC protein binds pseudo- or non-topologically with sufficient rate modulated by friction between the protein and fibre.

Key words: *DNA, chromatin, loop extrusion, SMC, cohesin, MD simulations*

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In vitro testing of a cathepsin K inhibitor as a potential therapy for osteoporosis

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The aim of this study was to determine the most appropriate concentration of anti-osteoporotic drug for the development of a new strategy for the treatment of osteoporotic fractures. The primary function of the drug is to inhibit cathepsin K (CTSK), the enzyme responsible for bone resorption, produced by osteoclasts, while promoting bone formation and mineralization. In *in vitro* experiments, the effect of three concentrations of CTSK inhibitor on human and rat osteoclasts was observed compared to a control without added inhibitor on cell metabolic activity and CTSK activity. Osteoclasts were obtained by culturing mononuclear cells from peripheral blood, in the presence of growth factors M-CSF and RANKL. To assess the cytotoxic effect of the drug on cells responsible for bone formation, the metabolic activity of the cells and the alkaline phosphatase (ALP) activity of osteoblasts obtained from rat femurs and the SaOS-2 cell line were monitored. Finally, the drug was tested on a co-culture of rat osteoblasts and osteoclasts, where both the metabolic activity of the cells and the activity of CTSK and ALP were evaluated. Human osteoclasts responded to the presence of a CTSK inhibitor with a slight concentration-dependent decrease in metabolic activity, which may be related to a similar trend in the decrease in CTSK activity. Rat osteoclasts responded with a statistically significant decrease in metabolic activity using the highest concentration of CTSK inhibitor, with a slight decrease in CTSK activity observed compared to control. In rat osteoblasts, cell metabolic activity was maintained at a similar level in all groups, while ALP activity was slightly increased when a CTSK inhibitor was added to the cells. Neither the metabolic activity nor the ALP activity of the SaOS-2 cell lines was affected by the addition of the CTSK inhibitor. The metabolic activity of rat osteoblasts and osteoclasts with the addition of a CTSK inhibitor to the co-culture was significantly lower compared to the control and with the number of culture days in the presence of the drug, we also observed a decrease in metabolic activity. CTSK activity decreased significantly compared to the control with an increasing number of culture days, which in turn may be related to a decrease in metabolic activity. Alkaline phosphatase activity was affected only in the presence of the highest drug concentration on the last day of culture, where significantly lower values were recorded compared to the control. These results suggest that the drug concentrations used may adversely affect osteoclast viability and thereby reduce CTSK production by osteoclasts. In further experiments, concentrations and culture conditions will be optimized to achieve an optimal drug concentration that will not adversely affect the metabolic activity of osteoclasts, but still reduce cathepsin K activity and promote bone cell proliferation.

Key words*: osteoporosis treatment, cathepsin K inhibiton, osteoclast, osteoblast*

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Stress-sensitive miRNA-based markers application for flax genotypes screening in the context of lignans content

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Naturally occurring lignans are present in seeds, nuts, cereals, vegetables and fruits. Flaxseed is the richest source of lignans. Lignans in plants are involved in plant defense mechanism through their antioxidant, antiviral, antibacterial and antifungal properties. Their content may be affected by a variety of factors such as genotype, tissue type, geographic origin, local environmental conditions, nutrition, and plant maturity. Genotypes of linseed, Czech (4) Dutch (3), French (1) and Canadian (1) origin were selected for the purpose of research into the potential impact of lignans on the degree of environmental adaptability of flax. According to the lignan content in the form of secoisolariciresinol diglycoside (SDG in $mg.kg^{-1}$), these genotypes are characterized by high, medium and low lignan content. Genomic screening was performed by touchdown PCR and application of molecular markers based on microRNA sequences 7-days-old seedlings were used to performed the screening by application of stress-sensitive miRNAs markers (miR156, miR168, miR167, miR172, miR395, miR396, miR398, miR399 and miR408), of which the following miR156, miR168, miR172, miR395 and miR408 provided positive amplification. The corresponding sequences of these markers at the level of miRNA molecules regulate the processes of homeostasis and accumulation of nutrients (phosphate, sulfur), tolerance to oxidative stress, biosynthetic processes, as well as growth and development processes. From the generated miRNA-based DNA loci profiles, the stress-sensitive marker miR408 was identified as a marker with the potential to differentiate individual genotypes at the polymorphism level, which was subsequently confirmed by genomic screening of seeds of nine flax genotypes. Preliminary results indicate a higher proportion of stress-sensitive miR408 marker sequences in the genome of high lignan genotypes.

Key words: *lignans, abiotic stress markers, miRNAs, polymorphism, flax*

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SECTION

Nutrition

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An assessment of ten menus of kindergartens in Slovakia

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Children nutrition is a notable topic with a big impact on future lives and the health of children. For their development and growth is necessary to fulfill the nutritional needs of children, to provide them high-quality food, balanced and varied meals. Numerous studies suggest that preschool age is key to building the right habits for a healthy lifestyle because at this age children start to be actively involved in making decisions about their behavior and they repeat what they can see around them. Preschool children usually spend at least half of the day in kindergarten, so school catering facilities have an important role in the fulfillment of mentioned needs. The aim of the study was to analyze and evaluate menus of two weeks from 10 state kindergartens chosen from whole Slovakia. In the present study, it was checked a compliance with the principles of rational diet, following the rules of slovak legislation set by The Ministry of Education, Science, Research and Sport of the Slovak Republic which is responsible for school caterings, and sufficient variety and diversity of menus. The frequency of serving of specific raw materials from food groups such as meat, fish, milk and dairy products, fruit, and vegetable or legumes was also counted. The relatively significant found feature is repeated serving of the same or similar meals, non-innovation of meals, the frequent serving of pastry or bread at the expense of other sources of carbohydrates, or frequent serving of sweetened drinks. In general, serving of fruit and vegetables could be more often. Just 3 out of 10 kindergartens served vegetables in addition and in sufficient amount on the daily basis (not just like ingredients in soup, sauce or other meals). Just 2 out of 10 kindergartens served dairy products daily. In one case fried food was offered, which is forbidden to serve in kindergartens by slovak legislation. To sum up the diet offered in the kindergartens is usually sufficient but with space for enhancement - a higher diversity could be present, dairy products, vegetables and fruit could be more often served and sweetened drinks could be exchange for tap water.

Keywords: preschool nutrition, kindergarten menu, food frequency serving, school catering

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The effect of oleic acid on acetone and urea in milk of dairy cows

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Supervisor: prof. Ing. Branislav Gálik, PhD.

High – yielding dairy cows needs the highest level of energy in feed intake in early stage of lactation. Energy is being mainly used on milk production. Metabolic disorder problems start, when level of energy in feed ratio is too low. Organism starts using energy from own body through the lipomobilization accompanied by higher level of circulating ketone bodies (OA, BHB, acetone and others) in blood. Level of oleic acid (OA), acetone (AC) and urea in milk can be used like one of the significant indicators of subclinical form of ketosis. The aim of the research was to analyze the effect of OA on acetone and urea in milk of dairy cows to 200 lactation days. Research lasted three years (from 2019 until 2021), In total 11 654 data were analysed. Three farms located in the northeast of Slovakia were included in this evaluation: 1st farm - 414 animals (60% Siemental cattle, 40% Holstein), 2nd farm – 275 animals (100% red Holstein) and 3rd farm - 517 animals (100% Siemental cattle). Farm 2nd didn't add any extra energy source to TMR, in farm 1st and farm 3rd glycerol had been incorporated. Data were available from breeding information system at the Ketosis report, and were subsequently sorted and evaluated statistically. Milk samples were analyzed by Slovakia Breeding Services. Statistical evaluation of data was accomplished in the SAS program. Based on concentration of oleic acid in milk the following groups of samples were proposed: A (0% – 0.5%), B (0.51% – 1%), C (1.01% – 1.5%), D (1.51% – 2%) and E (2.0% and higher). On each farm we monitored the highest level of AC in group E (2.0% and more), (1st farm 46.52 mg.l^{-1}, 2nd farm 47.22 mg.l^{-1} and 3rd farm 38.45 mg.l^{-1}). With increasing AO the AC increased in 1st farm (11.76 mg.l^{-1} – 46.52 mg.l^{-1}) and 2nd farm (18.31 mg.l^{-1} – 47.22 mg.l^{-1}), only on 3rd farm we monitored temporally decrease of the AC concentration in the group D (28.84 mg.l^{-1} – 17.06 mg.l^{-1}). Urea was highest on 1st farm (38.69 mg.100 ml^{-1}) and 3rd farm (36.01 mg.100 ml^{-1}) in the group E (2.0% and more). On 2nd farm urea reached the highest value (33.16 mg.100 ml^{-1}) in group D (1.51% – 2%). In conclusion, it is possible to emphasize the justification of the impact of nutrition quality with importance on energy feed intake in the first weeks of lactation.

Key words*: dairy cows, milk, oleic acid, acetone, urea, subclinical ketosis*

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The lockdown effect on schoolchildren – distance learning as a risk of childhood obesity onset

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Supervisor: prof. Ing. Marta Habánová, PhD.

Childhood obesity is a scarecrow of today's modern age, and the number of overweight and obese children is increasing by the year. Pandemic measures have worsened this situation and we now face an even more comprehensive challenge to address the deteriorating situation. The aim of the study was to identify changes in the body compositional status of children and their determinants created of anti-pandemic measures in Slovakia. We measured the body composition of 70 students (33 boys, 37 girls) aged from 11 to 17 years using bioelectrical impedance analysis (InBody S10). We focused on fat mass and fat-free mass indices. For tracking behavioral changes we used a questionnaire, and a comparison of the information from the questionnaire with the information of the body composition was made. Significant differences in body composition caused by lockdown in Slovakia were present in the visceral fat area, mean in 2020 was 49.49 cm^2 vs. the mean in 2021 was 52.44 cm^2 (p=0.037). The measured circumference of the abdomen also proved to be significant, the mean in 2020 was 73.88 cm vs. 75.40 cm in 2021 (p=0.003). In accordance, we observed longer sedentary behavior of children, caused by distance learning. Physical activity decreased due to the absence of typical Physical Education in schools. Changes in eating behavior, eating frequency, and drinking regime were present. The higher values of the measured circumference of the abdomen reflect the increasing visceral fat area. Accumulation of body fat in the abdominal area may onset many health risks in the future. To improve this emerging trend of larger fat storage it is necessary to educate children and parents about negative outcomes of such, possibilities of regulation, and improvement of body composition. Emphasizing physical activity to children is necessary, even more in the compulsory isolation period.

Key words: *COVID-19, lockdown, body composition, visceral fat area, waist circumference, dietary behaviors, physical activity, children*

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The mycotoxins content in corn silages with different additives

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Nutritive and fermentative value of silages can be improved by using various types of silage additives. It is well known that biological inoculants such as *Lactobacillus* have antifungal effect, and urea as a nutrient improves aerobic stability of a silage. Therefore, the purpose of this study was to determine the effect of a commercial inoculant *L. casei*, *L. buchneri* and urea, on mycotoxic contamination of corn silages. There were three versions of ensiled corn mass: variant C (control), variant A (inoculant addition) and variant U (urea addition). A dose of 1 $g.t^{-1}$ of inoculant and a dose of 5000 $g.t^{-1}$ of urea were added to the mass. After the treatment, mass was sealed in silage units and stored 12 weeks in the laboratory of Feed preservation at a constant temperature of 22 ± 2 °C. Mycotoxins content was detected by ELISA reader with the use of Veratox tests. Therefore, it was necessary to prepare the average samples according to established protocols. To determine the content of zearalenone (ZEA), fumonisins (FUM) and aflatoxins (AFL), an extract was prepared in 70% methanol, for ochratoxins (OTA) and T-2 toxin in 50% methanol and for deoxynivalenol (DON) in distilled water. The analysis showed a 100 % contamination of all corn silage samples. DON had the highest content of all mycotoxins regardless to the use of silage additives. ZEA, OTA and DON had higher ($P<0.05$) content after treating silage with inoculant (variant A). Compared to variant C, a lower ($P<0.05$) content of T-2 toxin and FUM was observed in variant A. Silage treated by urea had a higher ($P<0.05$) content of ZEA and a lower ($P<0.05$) content of T-2 toxin vs. control.

Key words: *corn silage, additives, biological inoculant, urea, mycotoxin contamination*

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Mucilaginous substances of flaxseed and their potential for functional foods

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Linseed flax (*Linum usitatissimum* L.) is classified as a functional food because of its nutritionally important content (Soni and Soni, 2020). Mucilaginous substances are found in the epidermal cells of the envelope layers of flax seeds, which are secreted after the seed is moistened. Their formation occurs in the Golgi apparatus, from where they are secreted into the apoplastic compartment via secretory vesicles (Naran et al., 2008; Western, 2012; Yang et al., 2012). The mucilage of flaxseed is composed mainly of water-soluble heteropolysaccharides, which, by their high molecular weight, represent about 3 to 9% of the total seed weight and are divided into two components: neutral and acidic. The neutral component is composed of D-xylose, L-arabinose, and D-galactose, while the acidic component contains L-rhamnose, L-fucose, L-galactose, and D-galacturonic acid (Kaur et al., 2018; Oomah et al., 1995). The mucilages in the plant perform several ecological functions, mainly they provide the seed with a moist environment, thus preventing it from drying out in dry environments and have positive effect to its metabolic activity and promoting its development. They also play a role in germination by adhering the seed to the soil, preventing seed dispersal or seed predation. They serve as a protective barrier against the external environment (Western, 2012; Yang et al., 2012). In the food industry, mucilage is important as a stabilizer, thickener and functional food (Basiri et al., 2018). As a functional food, it helps in digestive disorders, in the treatment of constipation where it acts as a laxative. It also helps in the treatment of obesity and has positive effects on high cholesterol, atherosclerosis and diabetes (Porokhovinova et al., 2017; Nybroe et al., 2016). The mucilage from flaxseed also exhibits antibacterial activity and also has potential in the treatment of chronic obstructive pulmonary disease (Mkedder et al., 2021; Khan et al., 2016). It has potential use in encapsulation of probiotics, e.g. *Lactobacillus acidophilus* La-05 (Bustamante et al., 2015). A survey of 200 linseed genotypes using 1.7 million SNP markers identified 7 quantitative trait loci associated with mucilage production. Candidate genes orthologous to *Arabidopsis thaliana* L. where they regulate mucilage synthesis and release, seed coat formation, and anthocyanin biosynthesis were characterized (Soto-Cerda et al., 2018). The knowledge of genotypic variability in the biosynthesis of mucilaginous substances will enable the identification of flax genotypes suitable for the creation of functional foods containing mucilage.

Key words*: flaxseed, mucilage, functional food, mucilage synthesis, candidate genes*

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Effect of the combined feed additive on the content of immunoglobulin A in colostrum and milk of sows

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For a new-born piglet, colostrum is a vital source of immunologically active substances that are important in protecting against potential infection. Immunoglobulin A (IgA) protects the piglets intestinal mucosa from pathogens during whole time of colostral and milk nutrition. The aim of this study was to assess the effect of the combined feed additive on IgA content in colostrum and sow milk. The combined feed additive contained a mixture of essential oils of oregano, anise and citrus and a prebiotic rich in fructooligosaccharides. The experiment included 14 pregnant sows of the Large white breed. The experiment was carried out in cooperation with the University agricultural farm VPP SPU Kolíňany, s.r.o. farm Žirany. There were 7 sows in the control and 7 sows in experimental group. The control group was fed a basal diet for lactating sows and the experimental group additionally received a combined feed additive at a dose of 1 g per kg of feed mixture, which started to be fed 14 days before the expected farrowing. Sows were housed in individual farrowing pens with a farrowing cage. The weaning was carried out on the 25th day, when the feeding of the combined feed additive was also completed. Colostrum and milk samples were taken by hand milking into tubes at 0, 12, 24, 48, 72 hours and on days 15, 20 and 25 from the birth of the first piglet. Subsequently, the samples were transported to the laboratory and frozen for further processing. Immunoglobulins were determined by an immuno-enzymatic method using a standard kit from Bethyl Laboratories, USA. Statistical analysis was performed using IBM SPSS v 26.0. Significant difference between control and experiment in IgA content was not recorded at any of the observed colostrum and milk collection times. However, positive trends in higher IgA content were observed in the experimental group at 24th and 48th hours and at days 15th and 25th. High variability of detected colostral and milk IgA within the same sampling time point was detected, which indicates considerable variability and differences in the concentration of IgA in colostrum and milk of sows. In the experiment, nonsignificant ($P>0.05$) positive effect of tested feed additive on the increase of IgA levels in colostrum and milk of sows was detected. However, further experiments are needed to confirm or refute the results.

Key words: *sows, immunoglobulin, nutrition, colostrum, milk, feed additive*

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Effects of antioxidant supplementation on the growth performance of pigs exposed to heat stress

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Heat stress (HS) is a great consequence experienced by pigs when global temperature rises due to climate change. Production parameters such as growth is highly affected by this condition as the animal will be forced to perform physiological mechanisms in order to reduce metabolic heat and maintain euthermia of which is an important factor for the animals' optimal productive performance. In line with the capacity of antioxidants (organic source) to alleviate the adverse effects of HS, it is this research's aim to assess the effect of antioxidant (vitamins C and E, and minerals selenium and zinc) supplementation at elevated levels on the growth performance of pigs reared under HS conditions. Thirty-six Danbred barrows (65 – 70 kg) were allocated into four treatment groups, one group placed in thermal comfort (TC) (20 °C) and the remaining groups (HC, HS1 and HS2) in HS condition (30 °C). The pigs were fed with their respective diet; pigs in TC received basal feed (B) and in HS room, diets B (HC), elevated 1 (increased levels of antioxidants, HS1), and elevated 2 (further increased in antioxidant levels, HS2) were given in *ad libitum.* Measurements were performed weekly for four weeks, which comprised the adaptation (7 days, fed B diet), gradual temperature rise (heat increment, fed experimental diet) in HS room (7 days) and the experimental period (14 days). Body weight gain (BWG), average daily gain (ADG), feed intake (FI), and feed conversion ratio (FCR) were measured. All throughout the adaptation and heat increment period, pigs in all treatment have similar measurements. HS had no significant effect ($P > 0.05$) on the BWG, FI and FCR of pigs, numerically, however, HC pigs had the lightest BWG and were the least efficient feed converter among the treatment groups. Significantly lighter ADG ($P < 0.05$) was observed in HC pigs in the 2nd week of the experimental period, whilst not statistically, slight improvement in the ADG of pigs were observed upon the supplementation of antioxidants at HS1 level. Our results indicate that, in certain growth performance parameters, pigs with high genetic capacity can be resilient to the adverse effects of HS, and for those parameters that is greatly affected by HS, it can be slightly improved by antioxidant supplementation.

Key words*: metabolic heat, euthermia, vitamin and minerals, production performance*

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Effect of the bee bread on the nitrogen profile of male Japanese quails (*Coturnix japonica*)

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Bee bread is characterized by its antibacterial, antiallergic, hepatoprotective, and antitumor properties, but has not been well studied yet. Nutrients from bee bread are well absorbed while digestion due to the process of fermentation. Legislation does not allow the addition of growth promoters to animal feed, therefore natural alternatives are being sought and bee bread might be one of them. The aim of this study was to evaluate changes in nitrogen profile by adding bee bread into feed of Japanese quails in different concentrations, which allows a better understanding of it´s possible effects. A total of 40 male Japanese quails (*Coturnix japonica*) were randomly divided into four groups according to administered bee bread into feed mixture HYD 11, which was given *ad libitum*, as follows: P1 (n = 10) 2 $g.kg^{-1}$ bee bread, P2 (n = 10) 4 $g.kg^{-1}$, P3 (n = 10) 6 $g.kg^{-1}$, and the control without additives (K) for 8 weeks. The groups were kept under the same conditions and parameters were analysed using an automatic clinical device Biolis 24i Premium. We noticed slight, not statistically significant ($P > 0.05$) increase in albumin in P3 (22.21 ± 15.51 $g.l^{-1}$) compared with control group (8.98 ± 2.99 $\mu kat.l^{-1}$), and in total protein between P3 (46.86 ± 18.27 $g.l^{-1}$) compared to P1 (28.32 ± 8.87 $g.l^{-1}$), not statistically significant ($P > 0.05$) as well. In treated groups we noticed slight not statistically significant ($P > 0.05$) decrease of uric acid in group P2 (125.3 ± 100.9 $\mu mol.l^{-1}$) and in the group P3 (154.3 ± 178.9 $\mu mol.l^{-1}$) compared to control group (284.8 ± 170.3 $\mu mol.l^{-1}$). Urea did not show any significant changes between groups. All of the parameters (albumin, total protein, uric acid and urea) were within the normal range. We can conclude that bee bread did not have a significant ($P > 0.05$) effect on the nitrogen profile by addition into feed, nevertheless, the results can be used for further examination. Despite the inconclusive results, bee bread might have positive effect on overall health by adding into the feed.

Keywords: *bee bread, beehive products, Japanese quails, nitrogen profile*

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Does increasing salmon intake help maintain vitamin D status in young Polish women in the autumn season?

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Background: Fish products are the main dietary source of vitamin D, but due to a low fish intake in the majority of European countries, an inadequate vitamin D intake is common. The matter is especially worrisome in regard to young women for whom it is essential for the osteoporosis prevention. Aim of the study: To assess the efficacy of a salmon intake intervention on maintaining vitamin D status in young Polish women during the autumn season, in which in Poland there is not enough sunshine exposure to generate skin synthesis. Material and methods: The dietary intervention comprised eight weeks of daily consumption of 50 g of Atlantic salmon and was conducted in a group of 47 women aged 20–30 years. Within the study, their changes of total serum 25-hydroxyvitamin D (25(OH)D) levels were analyzed and the effectiveness of the intervention depending on age, body mass index (BMI), and baseline 25(OH)D were assessed. Results: After 4 weeks of the intervention the median 25(OH)D decreased from 0.0571 mol/L to 0.0399 mol/L ($p < 0.001$). Nevertheless, after 4 more weeks of the intervention it increased to 0.0541 mol/L ($p = 0.001$), contributing to results not differing from the baseline ($p = 0.796$). At the same time, the share of respondents characterized by an inadequate vitamin D status increased until the 4th week, but afterward decreased until the 8th week ($p = 0.001$). Neither the age, nor the BMI influenced 25(OH)D during the study, whereas the baseline 25(OH)D was correlated with the BMI ($p = 0.042$; $R = -0.298$). The baseline 25(OH)D was associated with its levels during the intervention, as well as with 25(OH)D change from the baseline values ($p < 0.001$). Conclusions: In spite of the initial decline of the 25(OH)D observed, afterward the salmon intake intervention contributed to its increase.

Key words*: salmon, fish intake intervention, vitamin D, 25(OH)D*

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Evaluation of the differences between selected varieties of *Linum usitatissimum* L.

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Flax (*Linum usitatissimum* L.) belongs to Linaceae family and has been cultivated for fiber as well as functional food due to its potential health benefits and medicinal purposes. It contains compounds important in nutrition such as lignans or mucilage, which have antioxidant, antiviral, antibacterial and many other properties. The aim of this study was to evaluate the differences between varieties of oil flax. We used five varieties: Bethune, Astella, Raciol, Agram and Agriol. They were selected based on their composition- content of lignans, oils, fat, minerals etc. For assessment was applied phenotypic characterization using microscopic techniques. We randomly selected around 500 seeds from each variety to acquire images. In these images we evaluated the area of the object, the boundary height and width (Major, Minor), Feret's diameters (Feret, MinFeret), and Circularity. For image analysis we used specific program to process the images (open source software ImageJ version 1.53f51). The largest seeds were found in variety Raciol which was determined by traits – area, Major and Feret. The smallest seeds were found in variety Bethune. Using one-way ANOVA we confirmed statistically significant differences ($p \leq 0.0001$) between all analyzed varieties in the following traits – area and Feret. Between varieties Agriol and Agram were no significant differences in the trait Major. All other varieties were in this trait significantly different. In the trait Circularity was not found significant differences between the varieties Astella, Agram and Bethune. We can assume that the shape of the seeds of these varieties is similar. Through the used analysis, we determined that there are statistically significant differences in choosen traits between individual varieties. In the future we will identify individual phenotypic differences via further analysis.

Key words: *Linum usitatissimum L., flaxseed, phenotype differences, image analysis*

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SECTION

Multifunctional agriculture, environment, landscape architecture and rural development

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Endangerment of agricultural soil by potentially toxic elements along the Orava River

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Agricultural soil pollution by potentially toxic elements (PTEs) is a major concern nowadays. The pollution itself is usually caused by PTEs and their ability to enter the biotic parts of ecosystem, causing serious problems, eventually the risk for human health. In order to investigate the levels of contamination, pollution load index (PLI) and contamination factor (Cf) were calculated for the study sites along the Orava River. We selected 11 Fluvisols used as arable land along the River Orava, and two study sites located outside the alluvium (1 Cambisol used as a permanent grassland, 1 Technosol classified as an environmental burden). We determined basic chemical and physical soil properties, total PTEs content and the mobile fraction using inductive coupled plasma mass spectroscopy. The total PTEs content indicated their increased concentrations at many study sites. Chromium and arsenic were identified as the elements that contribute the most to the contamination of the investigated area, in which at least one limit value was the most often exceeded (total PTEs contents were compared with the limit values according to the national and foreign standards). From the point of potential food chain contamination, the most important are mobile and thus bioavailable fractions. The measured values of mobile PTEs fractions did not exceed the Slovak threshold limits in agricultural soils. Nevertheless, pollution indices reached increased values also in agricultural soils. According to the Cf, the Technosol was very highly polluted by Pb (12.89), Sb (11.94), Cd (11.82), Zn (9.19); considerably polluted by As (5.75), Cu (5.70), Mo (4.86); moderately polluted by Cr (2.47), Co (1.92), Ni (1.48). For Fluvisols, a moderate level of pollution was confirmed in the majority of cases. Average Cf for Fluvisols ranged from 1.02 to 9.00 for Cr, from 0.87 to 1.87 for As, from 0.76 to 2.37 for Pb, from 0.61 to 3.48 from Cd. In Cambisol we recorded low level of contamination for Zn (0.87), Cu (0.99), and Pb (0.94), and a moderate level of contamination for the others PTEs. The PLI reached in Technosol 6.02, in Fluvisols 1.46 (range from 1.08 to 2.09), and in Cambisol the lowest value (1.15). Despite the high values of the pollution indices calculated from the total contents, the mobile forms of PTEs were bellow detection limit. Based on the findings we can concluded that PTEs do not currently pose a risk to plant production. But the cumulative effect of total PTEs content can pose risk in the future.

Key words: *arable land, soil properties, potentially toxic elements, contamination factor, pollution load index*

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The environmental impact assessment of the inert waste landfill Podmaj on the environment and evaluation of its reclamation

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Current condition of environmental burdens in the country depends on its different methods of use, however the ultimate reasons of growing environmental burdens formations are anthropogenic, industrial and agricultural activities. Environmental burdens change some of stable and natural elements in the environment, straightly affecting biodiversity, pollution of the environment, risk of contamination and degradation of soils, ground water, surface water and atmospheric air. One of the evaluation impacts and risks of environmental burdens strategy is Environmental Impact Assessment (EIA). The aim of the research is to determine process EIA on the basis of the identified environmental burdens and to approximate and clarify the methodology of the Environmental Impact Assessment process. Process EIA describes the basic legal framework applicable in the process of assessing environmental impacts of environmental loads and it based on current laws, directives and regulations. The results of process EIA are opinions, processed and evaluated in expert opinions, which discuss the suitability of the proposed activities. The proposed activities are defined in the Atlas remediation methods for specific cases of reclamation and remediation of environmental burdens, which are coming out from the risk analysis of the affected area. The research work summarizes the brief characteristics of the natural conditions in the area of interest, which is territorial area Vrable and its current relation to environmental burdens. The main interest is devoted to assessing the impacts of the inert waste landfill Podmaj on the environment, where are evaluated its natural conditions, impacts on selected elements in the environment and its reclamation. The research work clarifies the natural conditions of the inert waste landfill Podmaj location, in the synthesis register, the degree of risk, location and type of activity that conditioned its origin, and also evaluates the operation of the landfill with respect to its location, which has significant landscape features. By assessing the impacts of the inert waste landfill Podmaj on the environment, it is possible to determine the EIA process, which also includes the assessment of reclamation, as an activity contributing to the successful elimination of the negative impacts of the inert waste landfill Podmaj. By adhering to the proposals and procedures, the reclamation of the inert waste landfill and its integration into the surrounding environment is a benefit for individual elements of the environment as well as landscape, and the goals of the process EIA have been met. Reclamation is a set of measures and interventions that mitigate or completely eliminate the adverse impact of environmental burdens in the country, which is also confirmed by the favorable results from the regular monitoring of these sites.

Key words: *Atlas remediation methods, environmental burden, Environmental Impact Assessment, inert waste landfill, reclamation, remediation.*

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Urban gardens in Ukrainian cities (Lviv, Kharkiv)

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Supervisor: doctor of geographical sciences, professor Nadiya Maksymenko

One of the popular areas of urban landscaping is the arrangement of urban gardens, which are accessible to the public. Urban garden is areas where people practice cultivation, mainly of food, in and around urban areas. Basically, it is the traditional cultivation of crops, but in urban centers. The idea of gardens and agricultural plots in the city is not new. In ancient times as well as the middle ages, farming was an integral part of city life and houses with gardens were a common feature of the city's landscape. Urban gardens are green infrastructure objects and provide environmental and social benefits. The growth of plants in urban areas comes in many forms and is influenced by various factors such as land space, topography, capital requirements, and the type of plants. Urban gardening can be done differently and includes aspects such as community gardens, urban farms, and aquaponics or hydroponics programs. In Lviv (Ukraine) the NGOs "PLATO", "Permaculture" and "EcoTERRA" include 1 hectare of land to the territory of the park "Zalizna Voda". The decision of renovated of public space of City gardening "Rering's Greenhouses" was made in cooperation with the city authorities and local residents. Previously, a large greenhouse farm was located in this area, which is in an abandoned state now. According to the project, this Center will provide services: growing organic products, mainly vegetables and fruits; conducting open lectures for the population; organization of a summer eco-cafe in which they plan to sell the grown products. For Kharkiv (Ukraine) is planning the arrangement of urban gardens in green zones (city's parks) near to housing zones (dense high-rise buildings). For example, it is proposed to create an urban garden in the parks "Zusrtich", "Zelenyi Hai", "Peremohy Park". The house building was created for workers of industrial enterprises who moved here from rural areas. It is this population that historically needs to realize the desire to work on land. The stages of preparatory works of urban garden include: 1. clearing of a site, garbage removal; 2. work to eliminate weeds; 3. construction beds for cultivation; 4. planting fruits and vegetables. Thus, such an urban garden will be the first such facility in these cities. Historically, only owners of private estates in Ukraine cities can have their own plots for growing. Urban gardens provide an opportunity to join the cultivation of organic products, planting, maintenance and collection of products for all citizens.

Key words: *urban garden, green infrastructure, landscape planning.*

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Conservation value of Orthoptera assemblages of a historically intensively used agricultural landscape: a case study in the Carpathian Lowland (East Hungary)

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Supervisor: Dr. Antal Nagy

The landscapes of European lowlands have been formed by agriculture in the last few thousands year, thus in these regions the main object of nature conservation is to protect the remain biodiversity of these secondary habitats. This diversity has outstanding importance not only in conservational, but also in agricultural aspect since yields depend on ecosystem-services. Ecological investigations mainly focus on the wildlife of remain natural habitats, and generally leave out of consideration the cultivated and other landscape elements covering especially larger part of the landscape. However, to understand the system functioning the investigation of these habitats (croplands, country roads and roadsides, etc.) is also necessary.
The Orthoptera assemblages of intensively used agricultural landscapes were studied in surroundings of Sajószöged along the Sajó and Tisza rivers (East Hungary), to evaluate which landscape element can preserve larger diversity compared to semi-natural control sites (pastures and hayfields). During our three-year study (2018 – 2020) samplings were carried out with sweep net and transect count methods in 36 sites representing all habitat types relevant to orthopterans. In the mainly unknown study area occurrence of 29 Orthoptera species (11 Ensifera; 18 Caelifera) were recorded, among which *Gampsocleis glabra* (Her.), *Acrida ungarica* (Her.) and *Celes variabilis* (Pall.) are protected. Based on multivariate analysis 4 types of Orthoptera assemblages could be defined. The intensively used croplands (maize, sunflower and wheat fields) had a less diverse and quite different Orthoptera assemblages than the less intensively cultivated lucerne and red clover fields, non-cultivated habitats (roadsides and country roads) and seminatural sites (hayfields, pastures) had. Composition and diversity of the studied assemblages mainly depended on the intensity of habitat use and secondarily on the culture. The grass like vegetation structure of less intensive lucerne and red clover fields preserved larger part of the natural fauna. Also the crop rotation served the maintenance of higher diversity. Landscape and cultivation planning using our results can provide sustainable agriculture that can maintain more diverse wildlife containing large number of beneficial (e.g. predator, and pollinator) and vulnerable species and species rich assemblages.

Keywords: *diversity, species composition, land use, cropland, vegetation structure, secondary habitats*

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Pre-treatment of lignocellulosic biomass by soaking in water to increase the biogas production

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Supervisor: prof. Ing. Miroslav Hutňan, CSc.

As lignocellulosic biomass is a material challenging to decompose due to its structure, this work deals with the study of a pre-treatment method promoting degradability of this material, and hence biogas yield therefrom. The studied lignocellulosic material is maize waste, which is one of the most common materials produced in agriculture. Using the correct pre-treatment method, this material could possibly represent a substrate for biogas production, as it would become a more accessible to anaerobic microorganism in the anaerobic digestion process. The investigated pre-treatment method is soaking the material in water, which could represent an economically advantageous and environmentally acceptable method used in practice. The test of absorbency of maize waste was performed to examine the effect of water on the dry material for a soaking time of 1 hour or for 1 day. Results of this test show that soaking the maize waste in water for the longer time decomposes the material more, which is expressed in a higher degree of solubilization. The average degree of solubilization for soaking time of 1 hour was 9.1% and 18.6% for soaking time of 1 day. Furthermore, for the study of this pre-treatment method, the long-term operations of two reactors for anaerobic digestion of such pre-treated maize waste were monitored. To one of the reactors the maize waste pre-treated by soaking in water for 1 day was dosed, while the maize waste dosed to the other reactor was soaked in water just before the dosing. During of 4 different phases of long-term reactors operations, production of biogas, methane content in biogas and changes in parameters of sludge water were monitored. During these phases the particle size of the maize waste, method of dosing the substrate and the volumetric load of the reactors were changed. The pre-treatment by soaking the substrate in water for 1 day had a more positive effect on biogas production compared to soaking substrate in water just before dosing, what is expressed by higher values of daily production of biogas, specific biogas production and higher methane content in biogas. By dosing only solids of the substrate soaked in water for 1 day, the highest value of specific biogas production (340.4 mL/g VS) was achieved.

Key words: *biogas, maize waste, methane, pre-treatment, renewable energy source*

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Development of the population structure of the red band needle blight origin in *Pinus nigra* plantation

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Supervisor: Mgr. Katarína Adamčíková, PhD.

Dothistroma septosporum and *Dothistroma pini* are two species of ascomycetous fungi of the genus *Dothistroma*, which causes red band needle blight disease, worldwide known as a Dothistroma needle blight. DNB attacks needles mostly of *Pinus* species and cause significant defoliation on their hosts. As both *Dothistroma* species produce nearly identical symptoms and have ability to cause same disease, the only unambiguous methods of distinguishing *D. septosporum* and *D. pini* is the application of molecular identification methods. PCR techniques involving 3 determination methods was used to compare the development of the population structures of both fungal pathogens of the two *Dothistroma* species, after 12 years at the same research area. The basis for this comparative analysis was the biological material we collected from the *P. nigra* plantation in 2016 and the collection of 24 isolates collected from the same research area in 2006 mentioned in the studies of Barnes et al. (2014). After the preparation of fungal cultures we have obtained single spore isolates that were used for DNA extraction from mycelium. Subsequent molecular identification determined of species transmitting DNB was realised by species specific primers using intronic regions of the β-tub2 and EF1-α genes developed by Ioos et al., 2010. MAT gene locus was identified by manting- type specific primers via amplified regions of both idiomorphs of the MAT gene by Janousek et al., 2013 and both mating type and species types at the same analysis was revelead using specific primers by Groenewald et al. 2007. From the whole number of 107 established cultures, we obtained DNA from 56 cultures, while the presence of both *Dothistroma* species was confirmed, however, 12 years ago only *D. septosporum* occured at the research area. For the species *D. septosporum*, we identified both mating types with slightly dominance of MAT1-2 and also simultaneous occurance of opposite mating types on single needle, whereas in the species *D. pini* we identified only the mating type MAT1-1. The close proximity of both mating types and their ratio 1: 1 increase the probability of sexual reproduction and the creation of a teleomorphic stage. These results suggest a potential increase in the genetic variability of the population and the possibility of adaptation of the fungus to the environmental conditions of Slovakia, while the occurrence of the sexual stage has not been confirmed in our country yet.

Key words*: DNB-Dothistroma needle blight, MAT -mating types, PCR techniques, DNA)*

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Impact of biochar application on CO_2 emissions and selected soil chemical properties

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Existing scientific research finds that biochar is a carbon-rich compound that generally improves soil structure and various properties. Also, biochar has potential to reduce CO_2 emissions from soils. Present experimental field study investigates the effects of biochar with or without nitrogen fertilizer (N) on CO_2 emissions and soil properties from Haplic Luvisol (HL) of Malanta site near Nitra in Slovakia. In this field study we evaluate the effects of biochar application on CO_2 emission and selected soil chemical properties during spring and summer season in 2019. HL was treated with biochar (produced by the pyrolysis of paper fibber sludge and grain husk at the temperature of 550°C for 30 minutes in Pyreg reactor) doses of 0 $t.ha^{-1}$ (B0), 10 $t.ha^{-1}$ (B10) and 20 $t.ha^{-1}$ (B20) in 2014. Each of these biochar amendments were combined with three levels (N0, N1, N2) of N fertilizer. The fertilization level of N1 was 108 $kg.ha^{-1}$ and N2 was 162 $kg.ha^{-1}$ in 2019. In treatments B0N0, B10N0 and B20N0, no N fertilized was used. Daily CO_2 emissions during study period depended on from air temperature. With higher air temperatures the concentrations of daily CO_2 emissions increased and with decreasing air temperatures, daily CO_2 emissions also decreased. It was found that cumulative CO_2 emissions from B10N0 and B20N0 increased by 1% and 8%, respectively as compared to its control B0N0. The cumulative CO_2 emissions from biochar treatments combined with fertilization level N1 also increased by 11% and 64% in comparison with control treatment B0N1. Opposite observation was found in case of B10N2 and B20N2 where the cumulative CO_2 emissions were reduced by 15% and 16% as compared to their control B0N2. The results showed that the application of biochar had a positive effect on the increase of soil pH, generally in all treatments. The mean concentration of NH_4^+ during study period decreased in all treatments as compared to control treatments. Statistically significant decreasing of concentration of NH_4^+ it was found after biochar application with or without N-fertilizer for treatments B10N2 and B20N2 in comparison with B0N2. Mean NO_3^- increased with addition of N fertilizer. The effect of biochar with or without N fertilizer increased SOC in the range of 2% to 65%. Correlations between average CO_2 emissions, soil pH, NO_3^-, NH_4^+ and soil organic carbon (SOC) were also studied. Significant correlation was found only between SOC and NH_4^+ (**$P<0.01$) and CO_2 and NH_4^+ (*$P<0.05$) measured in 2019. According to experimental results, it can be concluded that biochar with or without N fertilizer had a positive effect on all analyzed chemical properties in 2019. Also, biochar in combination with higher dose of N fertilizer decreased cumulative CO_2 emissions during study period. Therefore, biochar application produced by the pyrolysis of paper fibber sludge and grain husk could be considered in the mitigation of CO_2 emissions from fertilized Haplic Luvisol.

Key words: biochar, CO_2 emissions, soil chemical properties, nitrogen fertilizer

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Wastewater-based epidemiology as a tool for COVID-19 incidence in the population

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Supervisor: doc. Ing. Tomáš Mackuľak, PhD.

Monitoring of wastewater has emerged in recent decades as an effective tool for public health evidence by analyzing different chemicals, drugs, and pathogens in sewage. The presence of SARS-CoV-2 RNA in wastewater was first reported in March 2020. Since then, the potential of wastewater surveillance to contribute to COVID-19 mitigation measures has been further examined and incorporated in antiepidemic policies and strategies. Wastewater-based epidemiology is currently recognized globally as an early-warning tool to alert COVID-19 incidence. Tracking viral particles in wastewater may significantly help public health institutions to introduce relevant measures against the spread of the disease or to monitor effectiveness of the measures taken. Earlier identification of the virus presence in the population might limit the health and economic consequences caused by the next wave of the pandemic. Surveillance of the virus presence in wastewater can better estimate the COVID-19 widespread than regular direct testing, because wastewater monitoring covers the whole population including not-tested persons and individuals with only mild or no symptoms (major part of cases were asymptomatic). In our research, we investigated the prevalence of the COVID-19 disease in the population of Bratislava city, based on wastewater monitoring from September 2020 until March 2021. Samples were analyzed from two wastewater treatment plants present in Bratislava (Centrum –Vrakuňa and Petržalka) with reaching nearly 0.6 million monitored inhabitants. Wastewater analysis results reveal significant statistical dependence. High correlations between the number of viral particles in wastewater and the number of reported positive RT-qPCR tests of infected persons with a time lag of 2 weeks / 12 days ($R2 = 83.78\%$ / $R2 = 52.65\%$) as well as with a reported number of death cases with a time lag of 4 weeks / 27 days ($R2 = 83.21\%$ / $R2 = 61.89\%$) was observed. The obtained results and subsequent mathematical modeling provide a comprehensive early warning system for the identification of a local site of infection and, at the same time, estimate a possible pressure on public health systems up to two weeks in advance. Wastewater-based epidemiology in combination with direct testing of the population provides a comprehensive picture of the real situation in the country and thus significantly contributes to protecting public health.

Key words: *wastewater monitoring, epidemiology, SARS-CoV-2, COVID-19, public health*

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Weakness of strong ragweed

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Common ragweed (*Ambrosia artemisiifolia* L., Asteraceae) is an invasive alien species indigenous to North America. It has a negative impact on agriculture, biodiversity, and human health in Europe. The plant emits huge amount of pollen with the substantial allergenic potential, what is the cause of bigger losses as an economic damage to the agriculture. Ragweeds dominate in disturbed habitats and tends to spread rapidly and negatively affect populations of indigenous plants within ecosystems. The main goal of this study was to find out how to regulate pollen production of *A. artemisiifolia*. Ragweed was monitored in detail at three localities of Slovakia, Balvany, Malá nad Hronom, Veľký Horeš in 2019-2021. The occurrence of plants with special symptoms was evaluated. Typical symptoms were, young tissues pale - green/yellow, proliferation, malformation and virescence of male inflorescence, witches' broom, phyllody and plant´s stunting, but plants still stayed green/alive. The deformed male inflorescence forming "seeds" instead of pollen and whole plants were sterile. Production of pollen by symptomatic plants was assessed in the field. Investigation of 500 symptomatic plants confirmed that they did not produce any pollen. Extraction of DNA of symptomatic plants and subsequent PCR confirmed presence of phytoplasma. Phytoplasma is noncelular microorganism which cannot be transmitting by seeds. Phytoplasma need for its survival and spreading 2 hosts, plant, and animal. Animal vector is responsible for transmission of phytoplasma to the plant. To identify the presence of potential vectors, sweeping was used to determine potential vectors during June-August of each study year. One of the common transmitters are cicadas. By sweeping was captured 1800 specimens of cicadas. Among them was determined 46 species. The most promising vectors of phytoplasma could be three species from genera *Macrolestes.* In addition, another interesting vector was recorded within malformed generative organs of ragweed, Eriophyoid mite, which was determined as a species from genera *Aceria*. The Mites were recorded only from symptomatic plants, and they can transmit phytoplasma based on literature. It is a first record of such Eriophyoid mite from ragweed in Slovakia. This study suggests a weakness of strong ragweed – phytoplasma, which together with its vectors, can abolish pollen production and cause plant sterility.

Key words: *phytoplasma, cicadas, eriophyoid mites, excessive branching, male flower malformation, phyllody, sterile plants, pollen, witches' broom*

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Flower-visiting preferences of lepidopterans in Transcarpathian grasslands (Velyka Dobron' Game Reserve, west Ukraine)

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Supervisor: Dr. habil. Antal Nagy

About 80% of European plant species require insect pollination made mainly by bees (Hymenoptera: Apoidea) to produce seeds and fruits. Recent studies showed that beyond bees many other insect taxa such as butterflies (Lepidoptera), beetles (Coleoptera), hoverflies (Diptera: Syrphidea) can play important role in the pollination. Although many investigations were made on pollination activity of bees especially in case of honeybees and bumblebees, the role and importance of other taxa are less studied and known. Our study on butterfly pollination was made in grasslands of the Velyka Dobron Wildlife Reserve (West Ukraine). The flower visiting preference of species was registered along 100 m long transects. During the study data on 10 abundant butterfly species belonging 4 families and 26 abundant plant species belonging to 10 families were collected. Butterfly species visited different flowers with significantly different frequencies. The most visited plants were *Lythrum salicaria* (Lythraceae), *Betonica officinalis* (Lamiaceae), *Vicia cracca* (Fabaceae), while relative frequencies of butterflies was also higher than 10% also on *Centaurea jacea* (Asteraceae) and *Melampyrum nemorosum* (Orobanchaceae).

Keywords: *color preferencies, Hesperidae, Pierideae, Nymphalidae, Lycaendiea*

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Tree as a Gift: A Community-Supported Urban Landscape Project

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Supervisor: doc. Ing. Attila Tóth, PhD.

In 2013, the Municipality of Nové Zámky sold the public property in the built-up area of the town of 15 000 m^2 to a private developer for the construction of Baumax. Despite the negative attitude of the inhabitants and a petition against the construction, the fate of this area was irreversible. When seeing no will of the municipality to protect public green spaces, we came up with the idea to protect such places in the town. The community project 'Tree as a Gift' was initiated in 2017 by two experienced landscape architects – Slobodníková and Balogová with the help of their enthusiastic friends. We were looking for an effective tool to influence the municipality management through public spaces in the town. The best solution seemed through active civic engagement and direct participation on an emerging project. A project, where residents are not only final users of the space, but also its active co-designers under the supervision of experts. From the beginning, the emphasis was on a professional approach and outcome, even though the project aimed to be a voluntary and apolitical tool for the public to raise awareness and protect green spaces in the town. This project followed the philosophy of empirical learning, which can create a strong bond between people and place in the process of place making. We chose an area of 8 000 m^2 in the built-up area of the town. It was an unused, neglected space with the potential for transformation. We made a contract with the municipality to use this space for the purpose of planting and caring for trees. We started to promote the project to the public (FB page-600 followers) for a donation. The 'Tree as a Gift' initiative provides the residents with the opportunity to buy a tree, dedicate it to someone, and plant it in a designated public space designed by landscape architects. The planting was carried out in several stages, donated by different fundings. Altogether, 63 trees, and 510 shrubs were planted, and 132 m^2 of extensive perennial beds were established. During the planting, we discovered a hidden historical path called Mlynská Street with granite paving, which we used for constructing new and unique benches. A historical layer of the town came to live and become an important place-making element in the park. During the process a design was created and developed that one could not do from their computer desk, but only on site. The intention of landscape architects is to create the space with the public and at the same time educate them on the correct establishment and maintenance of different vegetation types. 'Tree as a Gift' is a pilot project that follows the methodology of research by design that can be tested, verified and implemented also in other public spaces for building a positive and strong relationship of public to their environment.

Key words: *Research by Design, co-design, participation, public open space, green infrastructure, landscape architecture*

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Comparison of spatial interpolation methods: Case study in Pripyat basin, Ukraine

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To create a continuous map of the distribution of hydrology characteristics over the insufficiently studied area, it is necessary to use various methods of spatial generalization. During this work was comparing the methods of Inverse Distance Weight (IDW) interpolation and Triangular Interpolation Network (TIN) on example of the Pripyat River basin within Ukraine. As a basic characteristic was chosen an average annual specific discharge, which in surface water hydrology means discharge per unit area of an upstream watershed. Specific discharge is highly usable for spatial interpolation, this characteristic is used to create cartographic materials and study insufficiently hydrologically unexplored areas. It was calculated for 31 hydrological observing posts. Pripyat river basin has insufficiently dense network of observation points and an uneven distribution of hydrological observing points within the basin. Despite a number of works devoted to the generalization of hydrological data within the Pripyat basin, full-fledged comparisons of different interpolation methods were practically not carried out. Analyzes of this type are conducted around the world, however their results can't be accurately extrapolated to other areas, which determines the relevance of this topic. According to the results of the work, it was determined that the IDW method is better suited for generalizations over the study area with a distance coefficient P = 5, the error of the obtained values is the smallest, the isolines are smoother, which better corresponds to the natural features of changes in the hydrological characteristics of the catchment. Checking with the help of hydrological posts not used in the creation of maps showed that on average the values taken from the maps differ from the real ones by 9-13%. The method of cubic TIN showed the lowest accuracy. In general, the TIN method in none of the variants can be used to accurately study the boundaries of the catchment and areas outside the interpolation. While IDW is also suitable for data extrapolation, shows good accuracy, and generally has greater prospects, TIN in these conditions can be used only to refine data in the central part of the right bank of the Pripyat, where the average errors of the method are lower than for IDW method.

Keywords: *Map creation, geographic information systems, interpolation, IDW method, TIN method, Pripyat*

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Nontarget catches of traps with synthetic attractants in northeastern part of the Pannonian Lowland

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Supervisors: Dr. habil. Antal Nagy, Prof. Dr. Miklós Tóth

The unique biota of the Nyírség and Hajdúság regions is known mainly on the basis of its flora, however, its fauna is poorly studied. Between 2013 and 2020 samplings were carried out in the southern part of the Nyírség and at 16 sampling sites of the Hajdúság during the development of synthetic (phenylacetaldehyde-based) and semi-synthetic (isoamyl alcohol-based) baits for pest monitoring, when many faunistical data on the Macroheterocera fauna were additionally gathered. In the south Nyírség 226 Macroheterocera species belonging to 7 familes, while in the Hajdúság 179 species belonging to the Sphingidae, Thyatiridae, Geometridae, Erebidae and Noctuidae families were caught. The selectivity and efficiency of tested baits differed notably considering both taxa (families and subfamilies) and abundance of catches. The semi-synthetic baits attracted more individuals and a wide range of noctuids mainly belonging to the Xyleninae and Noctuinae subfamilies, while synthetic lures showed specificity to species of the Plusiinae subfamily with lower abundances. Our study provided a large amount of faunistical data on mostly unknown Lepidoptera fauna of the region and brings attention to the alternative use of traps with synthetic attractants in faunistical studies.

Keywords: *Pannonian Lowland, phenylacetaldehyde, isoamyl alcohol, bait traps*

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Development of air temperature in the locality of water reservoir Turček

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In the context of ongoing climate change, a possible increase in the average air temperature of 1.1 to 5.4 °C is estimated worldwide under various scenarios, which could jeopardize the availability of water resources to the population. In Slovakia, the air temperature is estimated to increase by 2 to 4 °C by 2100. The paper will deal with the development of air temperature in the area of the Turček reservoir, which is intended for the production of drinking water for the population and its subsequent analysis using the Mann-Kendal statistical method, which is used to test trends, using levels of significance of – 0.1%, 1%, 5% and 10%, which are used to determine whether or not the data have a trend and, if so, whether they are significant or less significant up to insignificant. The studied locality is the Turček Reservoir, which is the fifth largest reservoir in Slovakia with a total volume of 9.9 million m^3 (the reservoir is filled twice a year) and was launched in 1996. The study contains results showing individual air temperature trends for each month of the year, during the period 2005-2019 as well as the overall trend for the whole period of study. As we can see from various published data in climate scenarios or in articles, air temperature is on the rise worldwide. This is also our case, although there are months in which the air temperature is even a declining trend (January, May, July and September). Nevertheless in the observed period 2005 – 2019, the total air temperature increased by 0.57 °C. The results of the Mann-Kendall test shows that only in one case it is possible to reject the null hypothesis without a significant trend and accept the alternative hypothesis, which means that there is a significant trend in July with significance level 10%.

Key words*: air temperature, trend, water reservoir*

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Antibiotics and their effect on activated sludge microorganisms

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The aim of this work was to determine the effect of three selected antibiotics - sulfamethoxazole (SMX), sulfapyridine (SP) and ciprofloxacin (CP) - on the activity of activated sludge (AS) microorganisms. For this reason, we performed several respirometric measurements. Respirometric measurements were performed in the presence of three different concentrations of these antibiotics. The sludge used in the measurements was taken from the wastewater treatment plant Bratislava - Devínska Nová Ves. First, endogenous respirometric measurements were performed to determine the effect of the monitored antibiotics on the endogenous activity of AS microorganisms. The percentage inhibition for these measurements ranged from 12.5% to 31.9%. The highest degree of inhibition was measured for the antibiotic SP (1.34 mg/L), while the lowest effect had the antibiotic SMX (0.142 mg/L). We also performed exogenous respirometric measurements using sodium acetate as an exogenous source of substrate. For exogenous respiratory rates, the inhibitory effects ranged from 1.13% (for SP at 0.134 mg/L) to 23.1% (for CPX at 1.04 mg/L). For substrate removal rates, the inhibitory effect ranged from 4.55% (for CPX at 0.104 mg/L) and 28.0% (for CPX at 1.04 mg/L). After performing respirometric measurements and their evaluation, we can confirm the expected influence of antibiotics on the activity of AS microorganisms, and thus on the activation process itself. Inhibitory effects were observed for all respirometric measurements and for all antibiotics monitored. This inhibitory effect of the antibiotics present in the activation process may indicate a deterioration in the ability of the AS microorganisms to remove dissolved forms of contaminants in the wastewater (if substances such as pharmaceuticals or drugs are part of the wastewater). It should be remarked that the values of antibiotic concentrations in these measurements were significantly higher than the real values of antibiotic concentrations in wastewater. However, due to the relatively high values of inhibition measured at our chosen antibiotic concentrations, we can assume a certain degree of inhibition even at lower antibiotic concentrations (but wastewater contains hundreds of pharmaceutical substances, the total concentration of which is often in the tens of thousands, sometimes in the hundreds of thousands of nanograms per liter) such as e.g., pharmaceutical concentrations present in wastewater. To clearly confirm these results, it is necessary to deal with the issue in the future.

Key words: *activated sludge, antibiotics, inhibition, respirometry, wastewater treatment.*

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SECTION

Plant Production

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Evaluation of molecular biological markers of different sweet corn (*Zea mays* L. convar. *saccharata* Koern) genotypes

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Supervisors: Adrienn Micsinai, PhD, Prof. Dr. János Nagy, DSc

Sweet corn (*Zea mays* L. convar *saccharata* Koern) is a popular vegetable all over the world. In the following years it is expected that research will focus on these varieties due to their important dietary role. Hungary has an outstanding record in the cultivation of sweet corn, being the largest producer in Europe and the world's second largest exporter - an average annual production of 500 thousand tons, 95% of which is exported. Plant carotenoids have been implicated in preventing several age-related diseases. Numerous studies have identified lutein and zeaxanthin to be essential components for eye health. They have also been linked with reduced risk of age-related macular degeneration (AMD) and cataracts, thus increasing the content of carotenoids in maize grains is of great interest. In our study we aim to follow the molecular biological markers of lutein biosynthesis by targeted gene expression studies in three super sweet maize varieties: Dessert R78, Messenger and Honey during the generative phase of the plants. At first, reliable reference genes will be identified for data normalization. Samples were collected from the topmost developed leaves of the plants, frozen immediately in liquid nitrogen and transported to the laboratory. RNA isolation included homogenization under liquid nitrogen, followed by RNA isolation, then it was transcribed to cDNA by random hexamer method. Real-time PCR was carried out on a Thermo QuantStudio 5 device utilizing SYBR dye kits. Previously we selected four reference genes for our studies, which encode tubulin (TUB), ubiquitin (UBI), actin (ACT) and a thioredoxin-like gene (TLG). These housekeeping genes consist of a group of constitutively expressed genes which are considered to be essential to maintain basic cellular functions, and are ubiquitously expressed in all cells of an organism, irrespective of tissue type, developmental stage, cell cycle state, or external signal. From these four candidate reference genes three (TUB, UBI and ACT) gave satisfactory results and selected for further downstream studies. The PCR products have also been confirmed by sequencing and sequence alignment to respective genes. For lutein biosynthesis gene expression, 7 target genes and 8 pairs of primers were selected for preliminary studies (PSY, HYD, CYP97C, PDS, ZDS, LCYB, LCYE). These indicated that all real-time PCR reactions are suitable for further studies in our selection of sweet corn varieties.

Key words*:* *Zea mays* L., *carotenoids, lutein, gene expression, housekeeping gene*

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Investigation of photosynthetic pigments in the fiber obtained from the wet fractionation of soybean (*Glycine max* L.)

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Supervisor: Assistant professor Dr. Szilvia Kovács

Preliminary studies on photosynthetic pigments have proved that in addition to their essential role in plants they also have benefits in humans and animals. Chlorophylls have antioxidant activity, antimutagenic activity, and induction of apoptosis in tumor cell lines. Carotenoids are able to bind free radicals and have positive effects on both animals and humans. Our present research aims to determine the quantity of photosynthetic pigments in fibers of different soybean varieties. The experiment was carried out in the summer of 2020 in the experimental garden of the University of Debrecen. Four soybean varieties were used, namely *Advisor, Bólyi 612, Isidor, Pannónia kincse*. Each variety was set up in 3 replicates and the plots were randomized in a block layout. One kg of green biomass per harvest was collected from each replicate, providing 3 kgs of sample per variety per harvest and we harvested two times in the season. After harvest, fiber was produced from the green biomass by wet fractionation. The basic step of wet fractionation is the separation of green biomass into two fractions by a mechanical press machine resulting green juice and fiber-rich fractions. The amount of photosynthetic pigments from frozen and lyophilized fibre samples was determined by spectrophotometric method. We applied variance analysis to make comparisons that resulted in a higher pigment content in soybeans sampled from the second harvest compared to the content of those harvested first. The highest amount of chlorophyll-a was measured for the Isidor variety (2.2771 mg/g dw) and the lowest for the *Bólyi* variety (1.7183 mg/g dw). In the case of chlorophyll-b, the highest amount was also in the case of the *Isidor* variety (1.1506 mg/g dw), the lowest in *Pannonia* (0.8968 mg/g dw). In the case of carotenoids, *Isidor* was again in the first place (0.8466 mg/g dw), *Bólyi* in the last place (0.6487 mg/g dw). In the case of xanthophylls, the highest amount can be detected in the case of the *Advisor* variety (0.3718 mg/g dw), the smallest in the case of *Bólyi* (0.3126 mg/g dw). We have found out that *Isidor* variety is outstanding among the four soybean ones investigated.

Key words*:* *soybean, fibre, photosynthetic pigment*

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Evaluation of NDVI, SPAD and nutritional values of sweet maize (*Zea mays*. L. convar. *saccharata* Koern)

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Due to the successful breeding and various innovative technological developments, the importance of sweet maize production and processing is growing on a global scale. Foods with high carotenoid content have positive effects on human health. Furthermore, research has shown that carotenoids in the human body can reduce the risk of developing age-related macular degeneration. Similarly to other researchers, Azami and Sun (2019) identified the antifibrotic, antioxidant, anti-inflammatory, antiapoptotic, antitumor, and chemopreventive properties of zeaxanthin. The experiments described in this paper were conducted at the Researcher Centre of the University of Debrecen. The super sweet maize Messenger (MES) was examined for its essential nutrient contents. In order to quantify carotenoid levels, the method of Moros et al. (2002) was used. Samples were dried at 50 °C and stored at 24 °C until processing to identify the various elements. Measurements were performed with ICAP 7000 spectrophotometer and the optimal one with no interference was selected, with the following spectral line overlap: Mg-285,213, P-177,495, Zn-213,856. Precision agriculture technologies were used during sowing, irrigation, fertilization, plant protection and harvesting. Sowing was performed on 22/05/2020 and harvesting was done on 17/08/2020. The amount of irrigation water was 88 mm and the applied fertilizer dose was 80 kg N, 21 kg CaO and 15 kg MgO per hectare. It was found that the measurement program described in this paper is able to provide complete information for the implementation of precision sweet maize production and production technology during the growing season. In addition, it is possible to determine the optimal sowing date. Furthermore, heat sum calculation can be used to monitor the development dynamics of maize, forecast silking, to identify critical events during the grain filling period based on precipitation and evaporation data, to plan possible interventions and to determine the optimal harvest time during ripening. During the content analyses of MES, high concentrations of phosphorus-, magnesium-, iron and zinc were observed (4455 mg/kg, 1530 mg/kg, 27 mg/kg and 27 mg/kg, respectively). As regards carotenoids, 9.57 mg/kg lutein, 8.93 mg/kg zeaxhantin, 0.725 mg/kg β-criptoxantin and 0.185 mg/kg β-carotin were measured, in addition to 2.45 mg/kg fructose, 2.6 mg/kg glucose and 3.675 mg/kg saccharose. Satisfactory and uniform NDVI values were associated with high yield quality. After the silking period, SPAD values showed close correlation with yield, which makes it possible to perform yield forecasting. During the growing season, the highest NDVI value (0.84) was measured before ripening, followed by a constant decrease.

Keywords: *sweet maize, carotenoids, mineral contents (phosphorus, magnesium, zinc)*

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Repellent effect of selected plant essential oils to the adults of *Leptinotarsa decemlineata* (Coleoptera: Chrysomelidae)

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Supervisor: prof. Ing. Ľudovít Cagáň, CSc.

The aim of the study was to find the potential repellency effect of two chemical substances DEET, 2-Undecanone, and essential oils (EOs) from *Allium sativum* L., 1753; *Artemisia annua* L., 1753, *Ocimum basilicum* L., 1753; *Lavandula angustifolia* Mill, 1768; *Eucalyptus globulus*, Labill 1800; *Pinus sylvestris*, L., 1753, *Melaleuca alternifolia* and *Curcuma longa* L., 1753, on the Solanaceae pest Colorado potato beetle *(Leptinotarsa decemlineata).* Olfactometer and Petri dish tests were used in the experiment. Five and three replicates were used for each tested dose (0.02, 0.04 and 0.06 ml/cm^2) of all EOs and two chemical compounds respectively. The average duration of one replication was 15 min. For bio-assay, 50 mg of cotton swabs were dosed with 10µl of the treatment solution and placed in one of the chambers of the olfactometer. The other arm received 10 µl of distilled water as a control. Ten adult beetles were released inside the base tube of the olfactometer. In the Petri dish test the average duration of one replication was 2, 4, 6, 24, 48, 72 and 96 hours. In every case, doses of each chemical and EOs were diluted to 10 µl of TWEEN 80 and dispensed on the surface of the filter paper. The control treatment contained a paper disk treated with 10 µl of TWEEN 80. The weight of the bait was 10 g of potato tubers as adult food. The petri dish was covered by the nylons to allow fresh air to move through. Five adult beetles were released in each Petri dish. The results showed that the greatest percentage of repellent effect of EOs in olfactometer test were detected for *A. sativum, O. basilicum, A. annua, P. sylvestris, C. longa, E. globulus, M. alternifolia* and *L. angustifolia* (89.7, 89.1, 88.9, 71.7, 68.9, 65.6, 58.4 and 53.5% respectively). On the other hand the Petri dish test detected the highest effect of *M. alternifolia, A. sativum, C. longa, O. basilicum, L. angustifolia, P. sylvestris, A. annua* and *E. globulus* (98.0, 92.8, 89.9, 83.9, 81.7, 81.5, 81.1 and 63.1% respectively). Our results indicate that all chemical substances and essential oils used in the experiment were repellent to *L. decemlineata,* in laboratory experiments. The study suggested other work to validate these results under field conditions.

Keywords: *repellents, plant essential oils, DEET, 2- Undecanone, Leptinotarsa decemlineata L.*

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Changes in qualitative and quantitative parameters of carrot roots as a consequence of the use of soil biostimulant Agriful

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Supervisor: doc. Ing. Miroslav Šlosár, PhD.

The research task was focused on an important type of root vegetable – carrot (*Daucus carota* L.), cultivar ´Romance F1´ (family: Apiaceae). The main goal was to verify the application of soil biostimulant Agriful (based on humic and fulvic acids) and its impact on the yield potential and quality of carrot roots. Nowadays, it is the common need to intensify and increase crop production because of growing human population and look for environmentally friendly cultivation methods. In general, biostimulants are biologically based products and their purpose is to stimulate the natural nutritional processes in cultivated crops. In realised experiment, Agriful (treatment of 5 l/300 l water/ha) was applied twice per vegetation period by spraying over the pivot. The increase of average root weight after the application of Agriful was found, concretely about +4.47% compared to the control variant. The higher root weight after Agriful application resulted in higher total yield of carrot about +2.84% compared to the control variant. Quality of consumable part of carrots were evaluated on the basis of the classification of roots into quality classes according to the valid standard for fresh carrot marketing (UNECE FFV-10). In the ´Extra class´, there was an increase in the average yield after the application of Agriful about +5.4%. The average carrot yield in ´Class I´ was decreased about -1.0% and root ratio in the ´Class II´ decreased about -1.9%. The ratio of ´Non-standard´ carrot roots was lower about -2.5%. Based on evaluating of qualitative substance content (total carotenoids, refractometric dry matter), the positive influence of the effect of Agriful application was found. The content of total carotenoids was higher about +8.7% compared to the control variant. The refractometric dry matter was higher about +4.1% compared to the control variant. The obtained results can be used in further research on biostimulants and it is possible to create clear recommendation for using of Agriful for small growers. It should be useful to verify these results in another vegetation period for recommendation to large-scale producers of carrot.

Key words: *biostimulant, carrot, yield, carotenoid, refractometric dry matter*

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Optimization of genomic DNA extraction from *Avena sativa* L. grains for the evaluation of variability of DNA amplicon profiles generated by Bet v 1 and Bet v 2 homologues

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Supervisor: prof. PaedDr. Ing. Jana Žiarovská, PhD.

For every molecular genetics study, it is essential to obtain high quality and pure DNA. The process of DNA isolation from plant tissues presents several problems. Plants contain of polysaccharides, pigments, phenols, secondary metabolites and their variation depending on plant species and plant tissues causes that there is no universal protocol for plant DNA isolation. Contamination such as secondary metabolites in isolated DNA samples can lead to inhibition of PCR reactions. In this study 4 available methods for the extraction of genomic DNA from oat grains were investigated: the well-known CTAB method, CTAB method with modifications such as adding PVP (polyvinylpyrrolidone) and DTT (dithiothreitol) to the CTAB buffer in the first step, and two commercial kits. The first and second method had similar results, the quantity of extracted DNA ranged between 8.50 to 244 ng. μl^{-1}, the quality measured spectrophotometrically (A260/A280) 1.30 and 6.34. The best results we obtained using the commercial kit EliGene® Plant DNA Isolation Kit (Elisabeth Pharmacon). DNA quality A260/A280 ranged between 1.99 and 2.11 and the quantity between 74.5 and 198 ng. μl^{-1}. The genomic DNA isolated with this method was used in the PCR reactions. Primers were chosen to match conserved regions of Betv 1 and Bet v 2 pollen allergens. The variability of 7 oat varieties (Zvolen, Valentín, Dalqes, Earl, Vendelín, Black beauty, Floron) was analysed with the mentioned primers. With the primer pair Bet v 1 forward and degenerated Bet v 1 reverse 49 amplicons were amplified in total. The length of the amplicons varied from 265 bp (base pairs) to 1929 bp; with the primer pair Bet v 2 (profilin) forward and Bet v 2 (profilin) reverse 61 amplicons were amplified in total. The length of the amplicons varied from the lowest 95 bp, which was shared by 6 of the varieties except of variety Earl, to the highest at 2432 bp, generated only by variety Black beauty. All varieties generated unique amplicons with both primer pairs. There was not detected an amplicon of specific size shared by all genotypes. The study also showed that primers for Bet v 1 and Bet v 2 can be used as universal markers for polymorphism in oat varieties.

Key words: *Avena sativa L., DNA extraction, Bet v1, Bet v 2, molecular markers*

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Evaluation of the effects of heat stress and acclimatization on the photosynthetic reactions of different wheat (*Triticum aestivum* L.) varietes

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Supervisor: doc. Ing. Marek Živčák, PhD.

Heat stress is one of the most important abiotic stressors that can limit the growth of cultivated plants in many regions of the world and can lead to a dramatic reduction in economic yield. The photosynthetic apparatus of crops is in direct contact with the environment and the processes of photosynthesis are almost immediately affected by any change in abiotic environmental factors. The aim of this study focused on the effects of heat stress, was to compare the reactions and resistances of varieties that differ from each other by morphological and photosynthetic features. Emphasis was placed on the measurement of photosynthetic and photochemical events measured by *in vivo* methods. The experiments were performed in the form of pot experiments. Long-term mild temperature stress (38 °C) was simulated by placing plants in a foil tunnel for 14 days, while short-term high temperature stress (45 °C) was simulated in a climate chamber for 24 hours. Fifteen genotypes were exposed to the effect of slightly elevated temperature, from which three genotypes were selected on the basis of leaf markings and photosynthetic manifestations, were exposed to the effects of short-term exposure to high temperatures. Previous exposure to slightly elevated temperatures allowed us to monitor the acclimatization reactions of selected plants when simulating the period of high temperatures. Structural and functional changes in photosystem II photochemistry were analyzed by rapid fluorescence measurements recorded with a Handy PEA portable fluorimeter (Hansatech, GB). The optical system MultispeQ v2.0 (PhotosynQ, USA) was used for multiparametric measurements of leaf signs and photosynthetic manifestations to obtain more detailed information about the physiological state and leaf signs. Slightly elevated temperature was able to stimulate chlorophyll accumulation in leaves, especially in mutant genotypes. The electrochemical shift (ECSt) was increased by temperature due to the increased accumulation of H^+ in the lumen of thylakoids. With increasing temperature, the permeability of the thylakoid membrane also increased, which was reflected in the increase of the proton conductivity gH^+. Comparison of genotypes allowed us to identify a significantly higher sensitivity of the mutant genotype ANK 32A to the effects of high temperature. However, the difference between genotypes disappeared when comparing plants that had previously been subjected to long-term acclimatization to elevated temperature. The increase in thermal resistance was probably associated with a significant increase in the parameter indicating photosystem I activity (ψREo). The results allowed us to identify genotypes with specific responses to thermal stress, which predisposes them to further research.

Key words: *photosynthesis, wheat, heat, stress, acclimatization*

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Identification of vicilin based amplified polymorphism in *Cicer arietinum* L. varieties

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Supervisor: prof. PaedDr. Ing. Jana Žiarovská, PhD.

Food allergies are becoming more common among different age groups nowadays, thus we can say that they are a today's modern age epidemic. Many times, people suffer from different health issues, caused by allergens in food. It is this fact that propels today's science in the study of allergens, which, in addition to proteomics, transcriptomics, also uses a genomic approach to research. For our research, we decided to choose the genomic approach, as we needed to identify the allergen vicilin in chickpea varieties, using generally designed primers. Chickpea is a legume with a balanced ratio of nutrients and in some countries, it is an irreplaceable part of the diet. For our research, we have chosen varieties which are large-scale cultivated but also regional varieties. Today, a lot of varieties are grown on a large scale, often for their high economic profit and not for their nutritional qualities. Vicilins belong to the cupin superfamily, which includes allergenic seed storage proteins. They are primarily localized in nuts, but also in legumes. They are stable to heat and proteases. In this study, we were aimed to the analyze of VBAP (Vicilin Based Amplified Polymorphism) in selected varieties of chickpea. VBAP is the newly designed marker technique that define the polymorphism of vicilin homologs that are the abundant part of many plant species. Vicilins have some positive qualities such as ability to act against fungal activities and microbials but can also cause an allergic reaction. The practical part of our research consists of isolation of DNA from frozen material using isolation kit GeneJET Plant DNA Purification Kit™ with some modifications. Firstly, the lysis buffers were replaced by 2x CTAB, second change were made in the incubation of the sample in thermomixer which lasts 50 minutes, lastly the precipitation solution was substituted by chloroform / isoamyl alcohol (24:1). The visualization of PCR products identified 3 amplified fragments (172 bp, 344 bp, 916 bp) with a 100% polymorphism among chosen varieties. For the phylogenic analysis we used UPGMA dendrogram method which was based on 0/1 binary matrix made of the amplified fragments. Based on the results of the dendrogram we can state the presence of 3 clusters.

Key words: *chickpea, DNA isolation, vicilin, allergen, polymorphism*

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Organic carbon and its labile forms as indicators of soil quality in selected systems of garden crops growing

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Supervisor: [1]Erika Tobiašová, [2]Ladislav Ducsay

The amount and quality of soil organic carbon is a crucial in the maintain of a healthy soil. In practice, it is necessary to react to changes immediately, and just the labile forms of organic carbon seems to be a suitable indicator of these changes in a short-time period. The experiment was located in the gardens of Slovak University of Agriculture on the soil type Eutric Fluvisol in the growing systems of strawberries (S) and cauliflower (C). Each of them included a control variant, variant with the application of a microbial isolate, variants with the application of AGRIFUL, GROUNDFIX and their combination, and finally a variant with the application of an inhibitor, while in all of which the DASA was applied. In the case of labile carbon oxidizable by $KMnO_4$ (C_L) after three months in both cases (S, C), its decrease compared to the initial content was determined. While in the case of C_L, its content was higher both at the beginning and after three months in variants with cauliflower, in the case of cold (CWEOC) and hot (HWEOC) water extractable organic carbon theirs contents were higher in the beginning in variants with strawberries, but after three months they levelled off. Significantly higher HWEOC contents were in the variants with the application of microorganism isolate and the lowest in variants with the application of GROUNDFIX or with DASA alone. Overall, in variants (S, C) with the application of GROUNDFIX there were recorded decrease in total organic carbon contents. In the variants with the application of AGRIFUL there were recorded increase, but only in the case of S variants. Whereas it is the total and not soil carbon, it was a decomposition of organic carbon originating from organic inputs, which is not bound to the mineral portion – particulate organic matter that means the labile fraction of carbon, as well. In the variants with the application of inhibitor there was an increase of organic carbon (S, C), which was more marked in the S than C variants. The changes in labile forms of organic carbon were manifested also in a short-time period, while the most sensitive reaction was in the case of HWEOC. The application of individual preparations significantly affects not only the quantity, but also quality of organic matter in the soil and thus the overall quality of soil.

Key words*: organic carbon, labile carbon, water extractable organic carbon, Eutric Fluvisol*

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Content of calcium, magnesium and sulphur in carrot roots depending on the amount of vermicompost in the growing substrate

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The impact of vermicompost on the content of calcium, magnesium and sulphur was determined in pot experiment realized in a vegetation cage in the campus of Slovak University of Agriculture in Nitra. The aim of the experiment was to determine effect the dose of vermicompost on the content of calcium (Ca), magnesium (Mg) and sulphur (S) and their dynamics during the vegetation in the roots of carrot (*Daucus carota* L. ssp. *sativus*). The variants contained 20 kg of substrate. In the variants was different ratio of soil to vermicompost: 20: 0 (var. 1); 9: 1 (var. 2); 4: 1 (var. 3); 3: 1 (var. 4) and 1: 1 (var. 5). Crop samples were analysed 96 and 136 days after sowing. The content of nutrients in plants growing in a substrate with a vermicompost does not have to be higher than of plants growing in a substrate without a vermicompost. Has been confirmed, that with increasing content of available nutrients in the soil substrate, their content did not increase in plants. The content of calcium and magnesium was the highest in the control variant in both sampling dates. The sulphur content in the roots increased due to the increasing dose of vermicompost. During the vegetation, the content of calcium in variants 1 and 2 increased, but due to the increased content of vermicompost, its content subsequently decreased. The magnesium content also increased significantly in variants 1 and 2, but the increasing content of vermicompost did not cause a decrease its content. The sulphur concentration increased in carrot roots between 96 and 136 days of vegetation. The findings in the present work are consistent with those of other authors, who show a strong antagonistic effect between K^+ and Ca^{2+} and also K^+ and Mg^{2+}. Synergistic effect has been demonstrated between NO_3^- and SO_4^{2-}. These findings should be considered when we develop the criteria for valuation the nutritional status of carrots based on the nutrient content in the roots.

Key words: *vermicompost, macronutrients, Daucus carota*

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Acclimation of buckwheat (*Fagopyrum esculentum* Moench.) genotypes of different provenance on drought stress

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Supervisor: prof. Ing. Marián Brestič, CSc.

Drought represents an important multidimensional abiotic stress factor, occurrence of which, as one of the climate change-impacts, rises. Within an effort for mitigation and adaptation we are looking for sufficiently resistant crops with high nutritional value and low inputs. Buckwheat appears to have features of such plant. This study aimed to determine changes of phenotype, photosynthesis and biochemy of different buckwheat varieties exhibited to three diverse watering regimes as the indicators of drought resistance. To realize this experiment there were selected six varieties of different origin: Russia, Poland, France, Canada, Zimbabwe and Bhutan. The experiment took place in The AgroBioTech Research Centre of the Slovak University of Agriculture in Nitra on the phenotyping platform PlantScreen. 108 plants were singly potted. 18 plants from each genotype were equally divided into 3 groups: control group watered in 80% soil water capacity (SWC), medium-stressed group watered in 50% SWC and strong-stressed group of 30% SWC. Measurements were performed via phenotyping platform and simultaneously by hand-held devices. The former includes Fluorcam imaging unit, RGB imaging unit and Hyperspectral imaging unit while manual measurements were carried out by digital chlorophyll meter SPAD 502 Plus (Minolta, Japan), chlorophyll fluorescence analyser Handy PEA Plus (Hansatech Instruments Ltd, United Kingdom) and PhotosynQ (United States). Gasometric method of assessment photosynthesis was performed by LI-6400XT (Licor, United States). Outcomes of PlantScreen showed modulating effect of drought on the plant area, height, perimeter, roundness, compactness and eccentricity of plants within each water regime as well as intervarietal differences. Photosynthesis of plants under conditions of 30% SWC tended to decrease although the watering in 50% SWC did not show significant effect in comparison with control group by French genotype La Harpe, Russian Panda and Zimbabwian PI 482597. In general plants responded to the drought stress by lower stomatal conductance, decreased carboxylation efficiency, decelerated electron transport and higher water use efficiency. Results of this experiment suggest that the genotype with origin in the aridest country among the examined ones (PI 482597 from Zimbabwe) behaves as the most adaptable and suitable to the drought-stress conditions.

Key words: *buckwheat, drought, acclimation, phenotyping, photosynthesis*

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Aspects of the production process of sunflower (*Helianthus annuus* L.) depending on different cultivation technologies

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Supervisor: doc. Ing. Ivan Černý, PhD.

In a small-plot field experiment was monitored the influence of the weather conditions of the year and selected hybrids of the sunflower on the yield and oil content of the seeds. The experiment was carried out in the year 2020 in the fields of the Research Centre for Plant Biology and Ecology in Nitra – Dolná Malanta. This area is included in the warm climatic region, with an average air temperature during the growing season of 16.3 ˚C and with total precipitation of 327 mm. The sowing was carried out with an 8-row seeder, with sowing at the level of the growing clip 0.70 × 0.22 m. The experiment included 14 hybrids of sunflower classified into 4 cultivation technologies. Conventional technology (Edison, SY Duomo, SY Chronos), clearfield (NK Neoma), clearfield plus (SY Bacardi, SY Gracia, SY Neostar, SY Nexus, SY Onesatr, SY Strato) and express sun (NX 92250, NX 92251, Subaro, Suffix). The average value of sunflower seeds yield was 3.62 $t.ha^{-1}$. In the monitored range of hybrids, within the individual types of cultivation technology, the highest yields, compared to the average yield, were recorded by SY Bacardi (clearfield plus) at the level of 4.17 $t.ha^{-1}$ (+0.55 $t.ha^{-1}$; rel. 15.10%) and by NK Neoma hybrid (clearfield plus) 4.06 $t.ha^{-1}$ (+0.44 $t.ha^{-1}$; rel. 12.14 %). In express sun technology, the highest yield was recorded by Suffix hybrid 3.72 $t.ha^{-1}$ (+0.10 $t.ha^{-1}$; rel. 2.64 %) and among the conventional hybrids is it SY Chronos with a yield of 3.58 $t.ha^{-1}$ (-0.05 $t.ha^{-1}$ rel. -1.26 %). The average value for the oil content of the seeds was 47.20 %. In the monitored hybrids, the highest oil content, compared to the average oil content, was recorded by SY Bacardi (clearfield plus) at the level of 49.41% (+2.21%; rel. 4.69%) and SY Duomo (conventional hybrid) 49.16% (+1. 96 %; rel. 4.16%). From express sun technology, was recorded the highest oil content by Suffix hybrid 47.01% (-0.19%; rel. 0.40%) and by the NK Neoma hybrid (clearfield) was the oil content 46.77% (-0.43%; rel. 0.91%).

Key words*: sunflower, production, hybrid, yield, oil content*

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Cultivation of chamomile and evaluation of selected elements of harvest and quality

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Supervisor: doc. Ing. Miroslav Habán, PhD.

Chamomile (*Matricaria recutita* L.) belongs to the family of Asteraceae, and is one of the most widely used medicinal plants in the world. It has many biological and pharmacological effects, such as anti-inflammatory activity, antioxidant properties, antimicrobial, anticancer, antispasmodic, and sedative effects. The aim of the work was to evaluate the production and selected compounds content of chamomile, cultivated in the company Agrokarpaty s.r.o. Plavnica, during the years 2016 to 2018. The subject of the research was the variety Lutea, which is characterized by a stable yield and higher essential oil content. In Slovakia, it is one of the most cultivated varieties of chamomile, and seeds for the experiment were obtained from a certified producer. The experimental site was located in a moderately cold, humid to the very cold climatic region at an altitude of 530 m above sea level. The predominant soil type of this area was cambisols. The same agronomic procedures were followed for all field experiments, and flowers of chamomile were harvested in the same period every year. The harvest during the monitored years decreased from 8.6 $kg.ha^{-1}$ to 7.3 $kg.h^{-1}$. By simple distillation of the drug of chamomile flower – Matricariae flos, the presence of essential oil in the amount of 0.68% in 2017 to 0.81% in 2018 was proved. For the analysis of phenolic acids (salicylic, chlorogenic and caffeic acid) the method of HPLC was used and subsequently, detection by DAD spectrophotometer. The year 2017 was dominant in terms of content substances. The amount of salicylic acid was 71.7 ± 3.68 $\mu g.g^{-1}$; chlorogenic acid 16.2 ± 1.6 $\mu g.g^{-1}$ and caffeic acid was 47.0 ± 4.1 $\mu g.g^{-1}$. The content of essential oils and phenolic acids was relatively balanced, which can be attributed to the genetic fixation of the constituents of the investigated chemotype of chamomile.

Key words: *Matricaria recutita, yield, chamomile flower, essential oil, phenolic acids*

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SECTION

Technology, quality and safety of raw materials and foodstuffs of animal origin

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Comparison of fat oxidation, protein degradation and sensory quality of carp (*Cyprinus carpio*) and rainbow trout (*Oncorhynchus mykiss*) meat after 24 hours post mortem

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Supervisor: prof. Ing. Juraj Čuboň, CSc.

The aim of the work was to analyzed oxidation stability and protein degradation of 3 basic market species in Slovakia. Samples were taken directly at the processing company, cooled in flake ice and shock-frozen (-36 ° C). The experiment included 30 fish divided into three groups. Group K (carp weighing 3 kg) 10 individuals, group PD (rainbow trout weighing 500 g) 10 individuals and group PL (rainbow trout weighing 2.5 kg). Samples from all individuals were taken at the place of slaughter for oxidative stability analysis (malondialdehyde content), total volatile basic nitrogen (TVB-N), raw meat texture (Warner-Bratzler shear test), meat color (CONICA MINOLTA 2600D) and sensory analysis of baked meat (color, tenderness, aroma, taste, juiciness). Analyzes were performed within 24 hours *post mortem*. The individual parameters analysed were processed in Microsoft Excel and GraphPad Prism 6 (Turkeys multiple comparisons test and basic column) on four evidence levels P - 0.05; 0.01; 0.001; 0.0001. The shear force value was highest in the PL group (0.1950±0.06525) compared to the K (0.1625±0.07206) and PD (0.1475±0.06089) groups; however these differences were not significant. The pH 24 hours post mortem was similar in groups PL (6.46±0.21) and PD (6.52±0.17) and differed in group K (6.35±0.11). The lightness L* was significantly higher ($P \leq 0.001$) in the PD group (54.00±2.453) compared to the PL (46.66±1.409) and K (50.80±2.997) groups. The reddest meat (a*) was observed in the PL group (17.58±1.743); the PD (0.7040±2.420) ($P \leq 0.0001$) and K (4.509±4.327) ($P \leq 0.01$) groups were significantly lower. The b* in the PL group (18.15±1.914) was significantly different from the meat of the PD ($P \leq 0.01$) (11.01±1.604) and K ($P \leq 0.001$) (10.40±2.546) groups. The content of malondialdehyde in meat of group K (0.4122±0.1480 $mg.kg^{-1}$) was significantly higher ($P \leq 0.001$) than in meat of PL (0.2454±0.2340 $mg.kg^{-1}$) and higher ($P \leq 0.0001$) than PD (0.1571±0.0506 $mg.kg^{-1}$). The content of TVB-N in the groups is PD (15.15±0.9650 $mg.100g^{-1}$) and PL (15.18±1.583 $mg.100g^{-1}$) is higher ($P \leq 0.0001$) than K (11.79±0.6254 $mg.100g^{-1}$). According to results of sensory evaluation, the PL (21.69±2.789) samples were in the first place before the K (20.88±2.167) and PD (19.88±3.389) samples. Samples from these species are further stored in the freezer and we will find out how the degradation metabolites and sensory friendliness to consumers will be assessed during long-term freezer storage.

Keywords: *fish, meat, fat oxidation, degradation, sensory evaluation*

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The impact of the level of hygiene in the process of honey production on the quality and safety of the final product

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We investigated microbial contamination of tools and equipment used to extract honey. From microbiological quality, we focused on the quantitative determination of the total plate count (TPC), the number of coliform bacteria (NCB), the number of filamentous microscopic fungi (FMF). After counting, the species of microorganisms were identified using the MALDI-TOF MS Biotyper. When evaluating the results, we divided the samples into primary (honeysuckle, pollen cell, empty cell, capped honey, honey cell and hive bottom), secondary contamination (watering can, a honey thrower, a hole for a honeycomb, a sieve, a small honey can, a large honey can, a shredding table, a shredding dispenser, a crowbar and a broom) and honey samples (rapeseed, agate, linden, facelial, sunflower and honeydew). We took samples in two different periods – June and September 2019. The average TPC value in primary contamination samples taken in June 2019 was 4.29 log CFU.cm^{-2}, in September 2.04 log CFU.cm^{-2}. The average TPC value in the secondary contamination samples taken in June 2019 was 2.60 log CFU.cm^{-2}, in September 1.85 log CFU.cm^{-2}. TPC in honey samples ranged from 1.67 log CFU.g^{-1} to 2.70 log CFU.g^{-1}. Coliforms bacteria were only present in samples taken in September 2019. The average NCB in the primary contamination samples was 1.75 log CFU.cm^{-2}. The presence of FMF in samples of primary, secondary contamination and honey was not confirmed. In samples taken in June 2019 we identified bacteria from the genera *Acinetobacter, Staphylococcus, Bacillus, Sphingomonas, Pseudomonas*. In samples taken in September 2019 we identified bacteria from the genus *Bacillus* and *Ralstonia*. The only bacterial spores that can survive in honey are the spores of *Bacillus cereus* and *Clostridium botulinum*. However, they cannot grow in honey and do not pose a danger to humans.

Key words: *honey, TPM, number of coliform bacteria, FMF, MALDI-TOF MS Biotyper*

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SECTION

Technology, quality and safety of raw materials and foodstuffs of plant origin

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Antioxidant activity, total polyphenol content and total flavonoid content of garlic (*Allium sativum* L.)

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Supervisor: prof. Ing. Judita Lidiková, PhD.

Garlic (*Allium sativum* L.) is a bulbous flowering plant belonging to the *Alliaceae* family and is one of the oldest traditional crops in the world. It is one of the most common types of vegetables and thanks to its characteristic aroma and taste, it is an important part of many world cuisines, and takes a significant place in the food industry. Although garlic is used mainly for its sensory properties, it is also known for its health-supporting qualities. Garlic has been used in the traditional folk medicine of many cultures. It has proven antiviral, antibacterial, antifungal, anti-inflammatory, and antioxidant effects. The health benefits of garlic are based on its chemical composition, which differs between cultivars and geographical areas but is also significantly affected by the growing conditions and cultivation practices. Garlic is characterised by a high content of polyphenolic compounds, mainly phenolic acids and flavonoids. Seven garlic cultivars, namely Arkus, Germidour, Karel IV., Makoi, Mojmir, Sukoradsky, Zahorsky were analysed in the presented work. Samples were provided by the National Agricultural and Food Centre – Research Institute of Plant Production Piešťany, Gene Bank of the Slovak Republic.This work aimed to determine the total polyphenol content, total flavonoid content, and antioxidant activity of selected garlic cultivars. Analysed parameters were analysed by the UV-VIS spectrophotometry method. Total polyphenol content (TPC) measured by Folin – Ciocalteau assay ranged from 430.26 to 640.04 mg GAE.kg^{-1} (gallic acid equivalent) FW (fresh weight). Total flavonoid content (TFC) measured by aluminium chloride assay ranged from 10.29 to 60.49 mg CE.kg^{-1} (catechin equivalent) FW. Antioxidant activity measured by ABTS assay (AA ABTS) ranged from 1.098 to 1.955 mmol TE.kg^{-1} (Trolox equivalent) FW. Antioxidant activity measured by FRAP assay (AA FRAP) ranged from 0.63 to 1.467 mmol.kg^{-1} FW. Statistical analysis was performed using RStudio software. Mojmir cultivar was characterized by significantly higher TPC, TFC, and AA. Zahorsky cultivar was characterized by significantly lower TPC and TFC. Sukoradsky cultivar was characterized by significantly lower TFC and AA. High positive correlations were determined between TPC and TFC (r= 0.94), TPC and AA ABTS (r= 0.78), TPC and AA FRAP (r= 0.87), TFC and AA ABTS (r= 0.77), TFC and AA FRAP (r= 0.86), and AA ABTS and AA FRAP (r= 0.94).

Key words: *Allium sativum; garlic; polyphenols; flavonoids; antioxidants*

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The effect of thermal processing on antioxidant activity and total polyphenols in sweet potatoes *(Ipomoea batatas* L.)

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Supervisor: prof. Ing. Janette Musilová, PhD.

Sweet potato (*Ipomoea batatas* L.) belongs to the most valuable food crop due to its high nutritional potential worldwide. Starch, protein, dietary fiber, vitamins, and minerals are the most abundant in sweet potatoes. Sweet potatoes are also rich in bioactive compounds, such as phenolic acids, flavonoids, anthocyanins, and carotenoids, providing a wide range of potential health-promoting effects (e.g., antioxidant, immunomodulatory, anticancer, antimicrobial, antidiabetic, antiobesity, and hepatoprotective). Polyphenols belong to the most abundant compounds in sweet potatoes. However, the content of phenolic compounds varies depending on the variety of sweet potatoes. Some differences in composition and the content can be detected also in particular morphological parts of the sweet potato root (flesh and peel). The most significant factors affecting the polyphenol content and bioavailability include the initial content in foods, food matrix, as well as food processing. In this study, the effect of heat treatment methods (microwaving, steaming, and baking), as well as the effect of sweet potato variety on the total polyphenol content and antioxidant activity of sweet potatoes, was investigated. For analysis, three sweet potato varieties grown in Croatia with different flesh colors – Beauregard (orange), O'Henry (white), and 414-purple (purple) were used. Total polyphenol content and antioxidant activity were determined spectrophotometrically. The total polyphenol content ranged from 0.53 (O'Henry) to 5.60 mg of gallic acid equivalents per gram of dry weight (mg GAE g^{-1} DW) (414-purple) in raw sweet potato (flesh) and from 0.98 (steaming – O'Henry) to 28.04 mg GAE g^{-1} DW (baking – 414-purple) in heat-treated sweet potatoes. The content of polyphenols was statistically significantly higher in baked sweet potatoes compared to other analyzed samples. The highest antioxidant activity was detected in steamed samples of variety 414-purple (4.51 and 19.57 µmol of Trolox equivalents per gram of dry weight (µmol TE g^{-1} DW) for DPPH radical scavenging activity and Ferric reducing antioxidant power assay, respectively). A strong relationship was found between both antioxidant activity methods. All studied heat treatment methods had a positive effect on the total polyphenol content and antioxidant activity of sweet potatoes.

Key words: *sweet potato, heat treatment, polyphenols, antioxidant activity, DPPH, FRAP*

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Impact of microwave radiation and plasma treatment on microbial quality of apple pomace

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Apples are the most cultivated fruit in the world and their processing is responsible for the production of large amount of waste. The result of apple processing is a waste product known as apple pomace. In view of the potential nutritional value of apple pomace, as well as the sustainability aspect and the financial demands for the disposal of apple pomace, apple pomace is considered for use as functional food product with a high nutritional value. As it is subject to undesired microbiological deterioration, it should be treated. There is little data available on the impact of apple pomace treatment on microbiological quality. The aim of our study was to treat the apple pomace using atmospheric plasma (experiment was performed in cooperation with Kamea Electronics s.r.o., Piešťany) and microwave electromagnetic radiation (Faculty of Biotechnology and Food Science, SUA, Nitra). We evaluated and compared the influence of both types of treatment methods on the microbiological quality of apple pomace. Using microbiological analysis, we determined coliform bacteria (KB), fibrous microscopic fungi and yeast (VMH and K). We analyzed totally 7 dry samples of apple pomace. We used the plate dilution method. Due to the fact that the dried samples showed the water activity lower than 0.95, we used DG18 agar as the culture medium for VMH and K determination and VRBL medium for KB determination. We monitored the given microbiological indicators before and after treatment of dry apple pomace (powder) with atmospheric plasma and microwave radiation (for 1 minute at 650W). Results of microwave treatment of apple pomace showed no presence of KB (<10 x 10^1 CFU.g^{-1}) in any sample before or after its treatment. VMH and K occurred before microwave treatment in sample no.4 (5.45 x 10^2 CFU.g^{-1}) and no.5 (9.10 x 10^1 CFU.g^{-1}). After microwave treatment in sample no.4, VMH and K decreased to 9.10 x 10^1 CFU.g^{-1} and in sample no.5 increased to 1.82 x 10^2 CFU.g^{-1}, in sample no. 7, an increase to 9.10.10^1 CFU.g^{-1} was found after irradiation. In other samples of apple pomace, VMH and K did not occur before or after microwave treatment. Before the treatment of apple pomace by atmospheric plasma we detected the presence of KB (2.52 x 10^3 CFU.g^{-1}) in sample no.5. After plasma treatment, KB were not detected to be in this (no.5) or in other samples (<10 x 10^1 CFU.g^{-1}). No colonies of VMH and K occurred (<10 x 10^2 CFU.g^{-1}) before or after plasma treatment of samples 3 and 7. Prior to plasma treatment, VMH and K were present in apple pomace in samples no.1 and 2 (9.10 x 10^1 CFU.g^{-1}) and in sample no.4 (1.64 x 10^3 CFU.g^{-1}). Samples no.5 and 6 were uncountable (<150 colonies). After treatment of these samples (no. 1 and 2) by plasma, no VMH and K colonies occured. In sample no. 4 the number of VMH and K colonies decreased to 8.18 x 10^2 CFU.g^{-1}, in samples no.5 and 6 they decreased as well. Based on our results, we can summarize that the treatment with plasma atmospheric discharge showed positive effect on the elimination of KB, VMH and K in apple pomace.

Key words *:* *apple pomace, microwave radiation, plasma treatment, microbiological quality*

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Jerusalem artichoke and its usage in baking industry

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Supervisor: doc. Ing. Ján Mareček, PhD.

The aim of this thesis was to prepare and evaluate cookies with sugar substituted with flour, made from jerusalem artichoke (*Helianthus tuberosus*, L.). We have prepared 4 samples – control sample (without jerusalem artichoke flour), and 3 samples with changed recipe with 33%, 66% and 100% substitution of sugar. We were analysing these parameters in the jerusalem artichoke flour and in the samples: dry matter, crude protein, content of fiber, crude ash, content of fat, content of polyphenols, content of flavonoids and phenolic acids, antioxidative activity and caloric value. Samples were also evaluated by sensoric analysis. We have found out that the jerusalem artichoke flour is a interesting product, because of its high content of fiber (4.82%), crude protein (11.72%) and crude ash (6.19%). Group of biologically active substances is dominated by the content of fenolic acids (1.27 mg CAE.g^{-1}). We have measured following parameters in 4 cookie samples: dry matter from 94.61% to 82.67%, crude protein from 8.60% to 11.67%, crude fiber from 2.64% to 3.4%, crude ash from 0.94% to 2.47%, fat from 19.11% to 18.71%, polyphenols from 0.31 mgGAE.g^{-1} to 0.45 mgGAE.g^{-1}, antioxidative activity from 1.74 mg TEAC.g^{-1} to 2.60 mg TEAC.g^{-1} and caloric value from 21.56 kJ.g^{-1} to 21.14 kJ.g^{-1}. The overall content of jerusalem artichoke flour in unbaked dough was 0% (control), 8.30%, 16.22%, and 22.81%. Parameters are written in order control – sample with 100% sugar substitute. Sensory analysis has shown, that cookies with higher sugar content were positively accepted by an average consumer, while cookies without sugar were not positively accepted, panelists concluded that they were bitter and crumbly. Cookies with 33% and 66% sugar substitute were accepted positively because of their nutty flavour. We can say that jerusalem artichoke flour is a good material for cookie production because it can add biologically important substances with antioxidative activity to the cookies, it can lower their caloric value and also add fiber to the diet of consumers, because it is known, that many people have low intake of fiber. Jerusalem artichoke flour can be a good substitute of sugar in pastry products, not so much for an average consumer but mainly for people suffering from some type of health or dietetary issues (for example *Diabetes mellitus*). Jerusalem artichoke flour can also be used to add nutty flavour to products, that do not contain nuts, which can be useful for people suffering from food allergies. Other factors should also be considered when preparing a pastry product recipe containing jerusalem artichoke flour, because jerusalem artichoke flour changes not only chemical and dietetary parameters, but also rheological and structural parameters of dough and finished product.

Key words: *Helianthus tuberosus* L., *cookie, flour, fiber, sugars*

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