Slovak University of Agriculture in Nitra

FACULTY OF AGROBIOLOGY AND FOOD RESOURCES

FACULTY OF BIOTECHNOLOGY AND FOOD SCIENCES

FACULTY OF HORICULTURE AND LANSCAPE ENGINEERING

Scientific Conference of PhD. Students of FAFR, FBFS and FHLE SUA in Nitra with international participation
-
Proceedings of abstracts

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

**7th November 2019
Nitra, Slovak Republic**

**Scientific Conference of PhD. Students of FAFR, FBFS and FHLE SUA in Nitra
- Proceedings of abstracts**

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Content

Preface

Considering the importance of science in the agriculture in our everyday life around us there is also the importance of these new knowledges and their possible interconnection.

Therefore, this conference Proceedings collected selected reviewed abstracts and provides an opportunity for students, scientists and other readers to engage with a selection of referred papers that were presented during the most comprehensive academic scientific conference of PhD. students hold on November 07, 2019 in Nitra, Slovak Republic. Submitted full scientific papers were reviewed and the best papers have been published in The Journal of Microbiology, Biotechnology and Food Sciences, Journal of Central European Agriculture, Slovak Journal of Food Sciences, Acta Fytotechnica et Zootechnica, Acta Horticulturae et Regiotecturae and Archives of Ecotoxicology. The scientific Conference of PhD. students is organised on the occasion of the Science and Technology Week in the Slovak Republic at the Slovak University of Agriculture in Nitra 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).

The main aim of the Conference was to provide a platform for presentation of research findings, exchange of experiences and knowledge sharing. The abstracts were split equally among the eight conference areas:

- Biotechnologies
- 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

There is no doubt that every our PhD. conference has encouraged the further advancement of interesting sources through fruitful discussions among students and other participants. Committees hope, that participants find valuable their engagement with various ideas in sustaining their own professional development and education; 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 as from the broader spectrum of biological sciences.

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SECTION

Biotechnologies

The effect of patulin on HT – 29 cell line

Rudolf DUPÁK

Supervisor: prof. Ing. Marcela Capcarová, PhD.

Patulin is a mycotoxin, secondary metabolite of microscopic filamentous fungi, produced mainly by *Penicillium expansum, Penicillium patulum, Aspergillus clavatus, Byssoclamys nivea* and *Alternaria alternata. Penicillium expansum* is the major pathogen responsible for patulin production. It is mainly found in apples, pears and their products. Patulin is characterized by a broad spectrum of toxicity, which includes both acute and chronic effects. The aim of our study was to observe effects of patulin on the HT – 29 cell line. The HT – 29 cell line consists of colon cancer cells with characteristic properties of the small intestine. The HT – 29 cell line serves as a suitable *in vitro* model for monitoring the effect of mycotoxins on the gastrointestinal tract. The study focused on observing the viability and antioxidant parameters of HT – 29 cells after the addition of patulin. Patulin was applied in concentrations: A (25 μg.ml^{-1}), B (12.5 μg.ml^{-1}), C (6.25 μg.ml^{-1}), D (3.125 μg.ml^{-1}), E (1.56 μg.ml^{-1}), F (0.78 μg.ml^{-1}) and G (0.39 μg.ml^{-1}). When monitoring the viability of the HT – 29 cell line after the addition of patulin, significantly reduced values ($P < 0.05$) were observed in the experimental groups A ($4.68 \pm 1.4\%$), B ($5.45 \pm 1.76\%$), C ($10.93 \pm 3.08\%$), D ($15.18 \pm 2.41\%$), E ($27.64 \pm 1.04\%$), F ($74.93 \pm 1.52\%$) and G ($92.38 \pm 2.59\%$) compared to the control group K ($100 \pm 0\%$), which did not contain mycotoxin. Superoxide dismutase activity of HT – 29 cells showed a significantly reduced difference ($P < 0.001$) in groups A (1.46 ± 0.085 U.ml^{-1}) and B (1.55 ± 0.071 U.ml^{-1}) against the control group (1.87 ± 0.035 U.ml^{-1}). Glutathione peroxidase activity showed both decreased and increased measured values, but no significant difference was observed in comparison with the control group (329.3 ± 10.11 U.l^{-1}). It means that patulin, at given concentrations, has no significant effect on glutathione peroxidase activity. This study demonstrates the negative effects of patulin at different concentrations on the HT – 29 cell line, due to a decrease in cell viability and the formation of oxidative stress, which can lead to other negative effects. Patulin is a highly cytotoxic mycotoxin that has reacted with the HT – 29 cell line at low concentrations. This study thus contributes to a better understanding of the effect of patulin and confirms its negative effect on the HT – 29 cell line. It is therefore important to monitor mycotoxin concentrations in order to prevent animal health threats, including threats in humans.

Key words: *mycotoxins, patulin, HT – 29 cell line, SOD, GPx*

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In silico characterization of the promoter sequence of the gene encoding β-1,3-glucanase

Monika FRÁTRIKOVÁ

Supervisor: Ing. Jana Libantová, CSc.

Sundew (*Drosera*) is a carnivorous plant which is characterized by high activity of hydrolases especially during digestion and defence processes, what makes this plant a perspective source of hydrolytic enzymes. Some out of hydrolases isolated from sundew can be used to increase plant resistance to phytopathogens using modern methods of biotechnology. It was shown that plant β-1,3-glucanase genes when they were introduced into transgenic plants, were able to enhance the plant defence against the microbial pathogens.

In previous study we focused on isolation of complete β-1,3-glucanase gene including the promoter sequence from. Here, we bring detailed *in silico* characterization of this regulatory sequence. Predictions of eukaryotic promoter and transcription initiation sites were performed using Neural Network Promoter Prediction software (http://www.fruitfly.org/seqtools/promoter). *In silico* analysis of the 5′ and 3′ untranslated regions for *cis*-regulatory elements with putative function was conducted using PLACE software (http://www.dna.affrc.go.jp/PLACE/signalscan.html/). The *in silico* analysis confirmed one teorethical transcription initiation sequence (TIS) and PLACE software recognized TATA box putative to promoter sequence 35 bp upstream from TIS. CAAT box element assumed to be responsible for enhancing of transcription was identified 59 nucleotides upstream of the core TATA box. Together we identified 11 copies of this element analysing both strands. Of the total length of 1154 bp of the promoter sequence, A+T pairs formed 62.1%, what is characteristic for plant promoters. Using PLACE program a number of regulatory motifs was explored and we subdivided them into four groups: light-responsive elements (GATA, I-BOX, T-BOX, CIACADIAN, GT1), tissue-specific elements (DOFCOREZM, CACTFTPPCA, RAV1AAT, ROOTMOTIFTAPOX1, POLLEN1LELAT52, GTGANTG10), hormon-responsive elements (ARR1AT, GAREAT, MYCCONSENSUALT) and biotic/abiotic stress-responsive elements (MYBCORE, ACGTATERD1, CURECORECR, WRKY-BOX, WBOXATNPR1).

Key words*: carnivorous plants, Drosera, β-1,3-glucanase, promoter sequence*

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Study of the influence of naturally occurring preparations DAG-1 and DAG-2 on oxidative processes of cotton in a saline environment

Sanjar NAVRUZOV, Ali AKHUNOV

Supervisor: Nigora Khashimova, DSc.

With the use of biologically active substances of preparations DAG-1 (a supramolecular complex of glycyrrhizin and salicylic acids) and DAG-2 (monoammonic salt of glycyrrhizic acid), can be used to control the genotype of plants the potential of molecular and biochemical adaptation to increase plant tolerance to stress factors.

The aim of the study is investigation the activity of antioxidant systems in adaptation to different salinization conditions of cotton, to identify the mechanisms of action of DAG-1 and DAG-2 on glycyrrhizic acid in soil adaptation processes.

Salt-resistant of C-6524 and sensitive C-4727 cotton seeds were selected as the study material. The seeds were soaked for 8 h in small concentrations of 10^{-7}M solutions of DAG-1 and DAG- 2 preparations, as appropriate for the natural phytohormones. The controls were grown in (without salt) aqueous and solution of NaCl in different concentrations (100 mM, 200 mM, 300 mM) (27 $^{\circ}$C) for 7 days. The amount of free proline was determined according to the Bates method. Malondialdehyde (MDA) scale was determined using thiobarbituric acid according to the instructions of V.V. Rogojin.

According to the results, salinity-resistant of C-6524 was accumulated a greater amount of free proline than the intolerant type of cotton. As salinity increased (in the order of increasing NaCl concentration), proline levels were correspondingly increased. As a result of DAG-1 and DAG- 2 drug treatment, the C-6524 grade was 11% and 52% in 100 mM medium, 29.4% and 17.6% in 200 mM medium, and 168% and 68% in 300 mM medium, respectively. In the C-4727 variety, proline levels were increased by 8% and 4% in 100 mM medium, 42.6% and 35.1% in the 200 mM medium, and by 107% and 20.4% in the 300 mM medium. It is well known from the literature that increasing of proline levels is considered a positive indicator.

Treatment with preparation DAG-1 of resistance cotton seeds C-6524 showed a decrease compound of proline 6.1% in controls grown by 100 mM NaCl medium, by 30.6% in 200 mM, and by 17.3% in 300 mM medium. The final product of the lipid peroxide oxidation process MDA was increased in all grown (100 mM, 200 mM, 300 mM) saline conditions of C-4727 non treatmented varieties, with treatmented DAG-1 preparation, in all salty conditions respectively decreased, 20% and 16.8% respectively. No statistical changes were observed in all samples under the influence of DAG-2.

It is assumed that the salicylic acid contained in DAG-1 stimulates the glutamate cycle, which facilitates the synthesis of proline amino acids, resulting in significant reduction in the peroxide oxidation of lipids. MDA accumulates poorly when plants are resistant to abiotic factors.

The prolonged and growth stimulating effect of the preparation was identified in pre-seeding treatment of cotton seeds in concentration of 10^{-7} M, comparable with the natural phytohormones, which enables to decrease the application ratio of chemical fertilizers and increase in cotton productivity.

Key words: *cotton, preparation, salt stress, proline, malondialdehyde*

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Influence of inoculant on content of microelements in selected types of legumes

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Supervisor: prof. RNDr. Alena Vollmannová, PhD.

Pulses are significant and still underrated crops. They are of great importance in the current diet in balancing the protein deficit of plant origin and in terms of nutrition they balance starch-rich cereals.

The aim of this work was to determine the influence of inoculant on the content of microelements in selected types of legumes. The evaluated material consists of 2 types of legumes - white lupine (*Lupinus albus*) and grass pea (*Lathyrus sativum L.*). Rizobine, which has a high content of live bacteria, was used as an inoculum before sowing.

We analysed dry seeds of legumes. Samples were analysed in two phases. In the first phase, the material was decomposed in a wet way with the addition of $10 \, cm^3$ of HNO_3. Mineralization of samples was performed by microwave digestion in the MARS X-press. In the second phase, after the mineralization, the residue was filtered and supplemented with distilled water in a 50 cm^3 volumetric flask to the mark to the exact volume. The analytical endpoint of the determination of the microelements content in plant material was the atomic absorption spectrometry on a VARIAN DUO 240F / 240Z instrument. All obtained values were calculated on dry matter content in order to assess the safety of monitored plant food raw materials as objectively as possible.

The Cu content in seeds ranged from 5.40 – 6.20 mg/kg for the lupine varieties and 7.40 – 8.80 mg/kg for the grass pea varieties in the inoculum-free variant. The addition of the inoculant increased Cu content in lupine varieties (6.24 – 7.30 mg/kg) and decreased it in grass pea varieties (5.30 – 8.10 mg/kg) compared to the control variant. Zinc in the varieties of lupine was in the control variant in interval 18.10 – 24.10 mg/kg, in grass pea 16.60 – 17.30 mg/kg. The addition of inoculant did not significantly change the Zn content in lupine (18.10 – 24.9 mg/kg), but slightly increased it in grass pea genotypes (15.50 – 18.70 mg/kg). The average Cr content in the control variant was 0.93 mg/kg in lupine, the addition of inoculant significantly reduced the chromium content to 0.44 mg/kg. The average Cr content in the control variant was 0.30 mg/kg in grass pea. The addition of inoculant did not significantly change the Cr content in grass pea 0.33 mg/kg. The nickel content in the control variant was in lupine (3.00 – 5.20 mg/kg). In variants with added inoculant the nickel content was (2.80 – 4.90 mg/kg) in lupine. The highest content of Ni in the pure variant was recorded in the lupine variety "Primorskij" 5.20 mg/kg and the lowest in the variety "WTD" 3.00 mg/kg. The average nickel content of the grass pea in the control variant was 1.73 mg/kg, and in the variant with the addition of inoculant was 1.57 mg/kg. The content of microelements in the monitored legumes also differed among species where the highest differences were recorded in the content of Fe. In varieties of lupine without addition of inoculant the Fe content was in interval 32.4 – 35.6 mg/kg and in grass pea varieties 36. 2 – 57.9 mg/kg. In variants with added inoculant the determined Fe in lupine and grass pea seeds were in intervals 32.10 – 39.20 mg/kg and 49.70 – 53.00 mg/kg (respectively).

Key words: *inoculation, lupine, grass pea, microelements*

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SECTION

Animal Production

Influences of initial body weight and biofilter type on inorganic nitrogen removal rates in recirculating aquaculture systems

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Supervisor: Prof. Miklós Bercsényi, and Prof. Ravi Fotedar

The study aimed to evaluate and compare the effect of initial body weight of fish and biofilter type on the nitrogen cycling efficiencies in recirculating aquaculture systems. Two sizes (9.4 ± 0.4 g and 107.4 ± 9.0 g) of koi carp *(Cyprinus carpio haematopterus)* were tested with two used biological filters; an aquatic plant (*Hydrocotyle rotundifolia*) and bacterial biofilms of a trickle-down filter in 12 independent recirculating systems for eight weeks. Each system consisted of three tanks: a fish rearing tank, a waste-collection tank and a biological filter tank. The efficiencies were evaluated by comparing fish growth, water quality parameters, and the removal capacities of total ammonia-nitrogen, nitrite-nitrogen and nitrate-nitrogen between the biofilters. The results showed that the body weight of fish did not affect the removal efficiencies of total ammonia-nitrogen, nitrite-nitrogen and nitrate-nitrogen, and both biofilters were independent of the effect of body weight. However, the bacterial biofilm filter showed higher removal rates of total ammonia-nitrogen and nitrite-nitrogen; while, *H. rotundifolia* had higher nitrate-nitrogen removal rates. The specific growth rate of koi was not affected by the type of biological filters; it was directly correlated with the effect of body weight. The specific growth rate decreases with increasing fish body weight; whereas the feed conversion ratio increases with increasing fish body weight. Despite, both biofilters were efficient in maintaining water quality parameters, and providing an acceptable environment for growth and survival of koi; an inverse relationship was observed between the initial body weight of fish and the total ammonia-nitrogen excreted into the fish tank.

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Increasing the accuracy of the estimation of breeding values of show jumping horse

Katarína DOČKALOVÁ, Juraj CANDRÁK

Supervisor: doc. Ing. Juraj Candrák, PhD.

The aim of this study was to analyze the variability (polymorphism) of genes NRAP, TRHR, NRF2 and MSTN in relation to show-jumping results and the possibility to increasea accuracy of breeding values estimation of selected population of horses in Slovak Republic. Horse performance testing is used as a traditional tool to predict a horse value as a breeding and sport horse in most warmblood riding horses in many countries. Horses are tested at a young age and the tests include conformation and performance traits. Heritabilites for many traits at tests are low to moderate. Therefore, these horses should be tested by new genomic methods which may lead to an increase in the accuracy of the estimation of the true genetic quality of the animals. We analyzed 116 horses in total. The analyzes included young horses 2 to 3 years old, horses participating in competitions 4 to 17 years old and breeding mares 6 to 23 years old. Genetic analyses were carried out for 5 single nucleotide polymorphisms (SNP), SNP1-BIEC2-1094761 for TRHR gene, SNP2 for myostatin, SNP3-BIEC2-689886 for NRF2 gene, SNP4-BIEC2-18316 and SNP5-BIEC2-84830 for NRAP gene. Relative Breeding Values for penalty points, for converted penalty points and for ranking were used. Relative Breeding Values were obtained from genotyped animals (full weight) or from genotyped parent horses (half weight) in case their offspring has not yet obtained his own breeding values. All breeding values were used for polymorphism analyses in relation to show-jumping results. We have found out that there is a connection between certain genotypes for given gene and Relative Breeding Values. The results of RBV averages showed that horses with genotype AA had the best Relative Breeding Values for TRHR gene. We are not able to confidently indicate which genotypes of horses are suitable for show jumping in regards to MSTN and NRF2 gene. Horses with genotype AA established for SNP4 BIEC2-18316 showed the highest Relative Breeding Values. Same similarity to horses with genotype TT established for SNP5 BIEC2-84830 was found. The statistical significance was confirmed for the influence of NRAP gene. By approximation the results of our analysis (increasing the values of the determination coefficients), we concluded that the inclusion of genotype information of selected genes increased the average accuracy of the estimated breeding values for horses by 6 % on average.

Key words: *genetic evaluation, breeding values, accuracy, show jumping, horses*

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Botanical origin of bee pollen loads in Slovakian apiaries

Ladislav KOHÚT

Supervisor: doc. Ing. Róbert Chlebo, PhD.

The aim of this study was to identify the main botanical sources of protein nutrition in Slovakian bee colonies. Pollen loads were collected from beehives at five locations across the country. In each location were selected three hives from which pollen loads samples were taken using bottom pollen traps. In nine dates, from April to September, pollen loads were sampled. The pollen loads samples were collected by using pollen traps mounted on bee hives entrances. In some localities there are missing some samplings due to personal issues of beekeepers or due to a bad weather. Each location was in different regions in Slovakia and in two kilometres area around each location types of landscape were defined. The locations were chosen to cover all area of Slovakia. Collected pollen samples were subsequently subjected to microscopic pollen analysis to determine their botanical origin. Most of the pollen presented in the hives environment comes from a relatively limited number of plant species and families. Only some plant families were determined as important pollen sources and only 11 plan families were the dominant pollen source in multiple localities. In early spring pollen originated mainly from willows, at the end of spring pollen originated dominantly from oilseed rape and where not available from plant families of *Aceraceae, Asteraceae* and *Rosaceae*. In the summer legumes and sunflower pollen loads begin to dominate and at the end of the season *Asteraceae* plants begin to dominate again, in mountain areas fireweed. Seasonal variability of floral resources in individual localities is analysed and impacts of sufficient supply of pollen on the bee colonies development is discussed.

Key words: *pollen loads, bee nutrition, pollen sources*

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Feeding grape pomace to horses: effect on digestibility of nutrients

Renata KOLLÁTHOVÁ, Branislav GÁLIK, Marko HALO

Supervisor: doc. Ing. Branislav Gálik, PhD.

The aim of the present study was to investigate the effects of dried grape pomace (DGP) supplementation on apparent total trackt digestibility of crude fat (EE), nitrogen free extracts (NFE), starch, sugar, non-fiber carbohydrates (NFC), crude fiber (CF), cellulose (CEL) and hemicellulose (HEM) in horses. The experiment was realized in cooperation with The Riding Centre of Department of Animal Husbandry (Faculty of Agrobiology and Food Resources, Slovak University of Agriculture, Nitra). Twelve adult Slovak warm blood sport horses were used. The digestibility analysis was carried out by total faeces collection (TFC) method. Nutrient composition of feeds and faeces was analysed in the Laboratory of Quality and Nutritive Value of Feeds (Department of Animal Nutrition, Faculty of Agrobiology and Food Resources, Slovak University of Agriculture in Nitra). Animals were divided into 3 groups: control group (without supplementation), group 1 (feed rations + 200 g of DGP) and group 2 (feed rations + 400 g of DGP). The addition of 200 g of DGP to equine feed rations showed a positive effect on the digestibility of CF, NFE and NFC ($P > 0.05$). Moreover, an increased EE and starch digestibility was detected at both levels of DGP inclusions ($P > 0.05$). The digestibility coefficients of sugar and HEM were practically not affected by DGP consumption ($P > 0.05$), however CEL digestibility have decreased in group 1, as well as in group 2 ($P > 0.05$). Although differences between the groups were not significant, these results suggest that DGP could be used in horse diets in the adequate dose for a possible digestibility improvement of some nutrients. However, to confirm the indicated positive trend for digestibility, further experiments with additional levels of DGP in horses are needed.

Key words: *digestibility, grape pomace, horses, nutrition*

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Different ways to compute genomic inbreeding

Kristína LEHOCKÁ, Radovan KASARDA, Barbora OLŠANSKÁ, Anna TRAKOVICKÁ, Ondrej KADLEČÍK, Nina MORAVČÍKOVÁ

Supervisor: prof. Ing. Radovan Kasarda, PhD.

The aim of this study was to describe and apply different methods of calculation of genomic inbreeding based on genome-wide SNP data in Slovak Spotted cattle. Totally, 85 animals genotyped using the IDBF International Dairy and Beef and the Illumina BovineSNP50 BeadChip were analysed. Traditionally, the inbreeding coefficient was estimated based on the pedigree data, but the development of new genotyping tools allows to evaluate the level of inbreeding at genome-wide level. Four methods were used to estimate the genomic inbreeding; analysis of runs of homozygosity (ROH) in the genome (F_{ROH}), identification of homozygous by descent (F_{HBD}) segments in the genome, calculation of the genomic relationship matrix (F_{GRM}) and the genomic relationship matrix from allele frequencies (F_{GOF}). Runs of homozygosity were identified in all animals. In the genome of Slovak Spotted cattle in average 82.91 ROH segments with length 58.81 Mb were detected. The distribution of ROH > 16 Mb showed that in current generation genomic inbreeding at level 0.44% can be expected. The F_{HBD} coefficient ranged from 0.15 to 0.35, depending on the HBD classes (1 to 10 K). The estimates of genomic inbreeding resulting from a genomic relationship matrix had negative values that indicated a relatively lower incidence of inbreed individuals in the evaluated population. On the other hand, the additive variance estimates equal to observed frequencies (F_{GOF}) indicated higher level of inbreeding (0.97). In general, the rate of inbreeding above 1% is recognized as a limit for the risk that the population in the long-term view will not survive.

Key words: *cattle, high-density SNP data, homozygosity, inbreeding, loss of diversity*

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Effect of *Lippia citriodora* extract supplementation on quality of rabbit semen *in vivo* and *in vitro*

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Supervisor: prof. Ing. Marko Halo, PhD.

Main component of *Lippia citriodora* extract – verbascoside is one of the most powerful free radical scavengers which exhibits a wide biological activity. During *in vivo* study, total of 20 New Zealand white rabbit bucks were split into two homogenous groups, one control (CON) and one verbascoside-supplemented (0.1%) in feed mixture (EXP) and later on *in vitro* analysis of verbascoside effect on rabbit spermatozoa motility aspects were executed. Parameters were measured using CASA method – automated microscopic spermatozoa analysis. Spermatozoa concentration, ejaculate volume, spermatozoa motility, progressive motility, distance parameters, velocity parameters and type of spermatozoa movement were all negatively affected by the extract of *Lippia citriodora* leaves after first four weeks of supplementation until the end of the experiment (8 weeks). All spermatozoa traits returned to normal values in line within control group after four weeks suspension of dietary treatment. For *in vitro* investigations, ejaculates of 10 male New Zealand white bucks were collected by using artificial vagina. Subsequently, samples were diluted in physiological saline solution containing different concentrations of verbascoside – 0, 0.0024, 0.0219, 0.157, 120 mg/ml (Ctrl, VB1, VB2, VB3, VB4 groups, respectively), using a dilution ratio of 1:4. Obtained data proved that verbascoside at concentration of 0.0024 and 0.0219 mg/ml has no adverse effect on spermatozoa. Furthermore, we found out that verbascoside at higher concentrations (0.157 and 120.0 mg/ml) significantly altered all the motility parameters analyzed in the experiment. To sum up a possible negative effect of verbascoside supplementation into feed mixture (0.1%) on semen quality parameters in rabbit bucks as well as *in vitro* can be stated, obviously considering that target organs of antioxidant activities of phenylpropanoid glycosides are various. In addition, it has to be emphasized that the extract showed a reversible action since the semen traits of treated animals returned to the normality after the dietary administration period.

Keywords: *Verbenaceae*, *food additive, spermatozoa, motility*

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Effect of calving season and temperature at calving on the gestation length

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The objective of the study was to assess the effect of calving season and the effect of average maximum temperatures during the last 6 weeks before calving on the gestation length of dairy heifers. Data, 393 records, were collected from three dairy farms located in the Slovak regions of Orava (Northern Slovakia), Upper and Lower Nitra (Western Slovakia), in the years 2006 – 2018. The herd "A" consisted of Slovak spotted breed, the herd "B" and "C" consisted of black Holstein Friesian cows. The heifers in the farm "A" and "C" were housed in free housing system without fans and in the farm "B" in the boxes also with no fans. In the herd "A" the housing conditions were suboptimal, because of it the ambient temperatures were significantly influencing conditions in the barn. Barns in the other two farms had ceiling. Mean 305-day milk yield in the herd "A" was 5529 kg, in the herd "B" 9402 kg and in the herd "C" 9655 kg per cow. The temperature factor was divided into four categories: below 5 °C, 5.1 – 14 °C, 14.1 – 20 °C and above 20 °C. In the farm "A" all four categories of the temperature were studied in the farms "B" and "C" only three categories were studied (below 5 °C was omitted). The observed factors were found in relation to gestation length insignificant. However, in our study we observed certain tendency, which might indicate some degree of effect of studied factors on gestation length. In the herd "A" we observed that heifers that were calving in winter had longest average gestation length (288.27 ± 2.32 days) and heifers that were calving in summer had the shortest average gestation length (280.78 ± 0.79 days). Similarly, to these findings we observed in this herd the longest gestation in animals that were exposed to the average maximum temperatures below 5 °C (289.75 ± 2.48 days) and the shortest gestation in animals that were exposed to the average maximum temperatures above 20 °C (279.62 ± 0.9 days). In the herd "B" the heifers that were calving in summer had the longest gestation (279.02 ± 0.42 days), in the herd "C" heifers that were calving in autumn (278.03 ± 0.61 days). The shortest gestations in the herd "B" and "C" were related to spring calving (275.98 ± 0.54 days; 276.22 ± 0.31 days). In conclusion, although our study did not reach statistical significance, we pointed out some tendency of relationship between season, temperature of calving and gestation length. Our findings from the herd "A" are consistent with several other authors, however surprisingly data from the herd "B" and "C" contradict them. Very low temperatures could be a factor causing prolongation of gestation length and high temperatures might cause its shortening.

Key words: *calving season, temperature, gestation, heifers, dry period*

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Genome-wide signals of artificial selection trough FST statistic in Slovak Spotted Cattle

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The objective of this study was to analyse the genomic regions that have been affected by artificial selection of Slovak Spotted cattle and other two breeds Holstein and Swiss Simmental, which were used for grading up of Slovak Spotted cattle in the past. In cattle, intensive selection for economically important traits is expected to have left selection signatures in the genome. The advances in genomics in the last years have enabled the development of several methods to detect selection signatures. The high-density SNP data of 85 Slovak Spotted, 99 Holstein and 78 Swiss Simmental were obtained. The standard quality control of SNP data was performed to exclude individuals with $> 10\%$ missing genotype across the autosomal loci, markers missing in $>10\%$ of individuals, and loci with minor allele frequency < 0.01 using the PLINK v1.9 software. After quality control, 35 934 autosomal loci with average spacing 70.22 kb were included in the analysis. The genome-wide scan for selection signals was performed trough Wright's F_{ST} statistic. The F_{ST} values were calculated for each locus using PLINK v1.9 software and then averaged over 5 consecutive SNPs using sliding windows approach adopted in the Zoo package. Top 1% of signals were recognized as outliers reflecting the regions significantly affected by intensive selection. The threshold value was set to 0.49. After applied approach, 19 genomic regions across 12 autosomes were identified to be under strong selection pressure. For each region with selection signals quantitative traits loci (QTLs) were identified. Directly in the signals on BTA 3, 5, 6, 7, 8, 12, 14 and 26 are located QTLs for milk production including milk fat yield, milk protein yield, and milk speed. On BTA 3, 4 and 10 were find OTLs affected beef production (marbling score, longissimus muscle area) and on BTA 2, 3 and 4 were identified OTLs mainly associated with fertility (calving ease, dystocia, age at first calving). Results showed that domestication and mainly artificial selection processes have definitely shaped the bovine genome. The identification of selection signatures in the genome allows to obtain new genetic markers that are involved in the genetic control of economically important traits and can be potentially used to genomic selection in the breeding practice.

Key words: *artificial selection, autosomal loci, genomic region, sliding window, Wright's statistic*

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Effective population size of Slovak Spotted cattle based on high-density SNP data

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The aim of the study was to analyse effective population size of the Slovak spotted cattle based on the linkage disequilibrium. Evaluation of the effective population size and linkage disequilibrium can provide important insights to understand the genetic diversity and population history of this dual-purpose cattle breed as one of the Slovakia landscape heritage. In total of 37 males and 48 females were genotyped using Illumina BovineSNP50 BeadChip and International Dairy and Beef chip, respectively. After SNP pruning, overall 37510 SNP loci were selected for the analysis of effective population size. The quality control was done by PLINK v1.9. The relationship between linkage disequilibrium (LD) and effective population size (N_{eLD}) was used to estimate the trends of recent and historical effective population size. The level of linkage disequilibrium, non-random association of alleles at two or more loci, was expressed across syntenic SNP pairs by parameter r^2_{LD}. The N_{eLD} calculation was based on its functional relationship [$E(r^2_{LD}) \approx f(c, N_{eLD})$] with r^2_{LD} and recombination rate (c) expressed as physical position (genetic distance) of syntenic SNP pairs in the genome (1 cM ~ 1 Mbp). The trend of effective population size was estimated using the SNeP v1.1 software. The results showed that the linkage disequilibrium decreased continuously with increased genetic distance between SNP loci. The trend of historical effective population size of Slovak Spotted cattle showed rapid decrease of N_{eLD} during last 60 generations, approximately 7.84 per generation ($r^2 = 0.99$). Due to the fact that the N_{eLD} estimates for first nine generations are biased, the recent effective population size (N_{eLD0}) was calculated by linear regression of N_{eLD} estimates for 10 to 60 generations ago. The analysis indicated that the long-term trend of genomic N_{eLD} is not positive and in current population of Slovak Spotted cattle only 32.76 individuals can be expected. Based on this, the Slovak Spotted cattle can be regarded as critically endangered by the loss of genetic diversity.

Key words: *cattle, genetic diversity, local breeds, SNP genotyping*

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Mastitis pathogens in sheep milk and their antibiotics resistance

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Supervisor: prof. Ing. Vladimír Tančin, DrSc.

The aim of this study was to find out the incidence of pathogens in sheep milk and determined their antibiotics resistance. The experiment was carried out at sheep farm during lactation period in 2018. In the study there were randomly selected 66 dairy ewes without symptoms of clinical mastitis. At sampling, the first squirts of milk were discarded, subsequently the udder halves were cleaned with 70% alcohol and approximately 5 ml of milk from each udder halves were taken in sterile tube for bacteriological examinations. Thus 132 milk samples were examined. The sample of milk was inoculated on the plates with 5% blood agar and were incubated aerobically at 37 °C and examined after 24 and 48 hours. Colonies were identified on base detailed reactions for detection of pathogens. Contagious pathogens (*Staphylococcus aureus, Streptococcus agalactiae*) were classified as positive if one or more colony-forming unit (CFU) were found. Other pathogens were classified as positive if at least five CFU were found out. Samples were classified as contaminated if three or more different pathogens were isolated from one milk sample and growth of contagious pathogens were not identified. Susceptibility testing was performed and evaluated according to the recommendations of CLSI (CLSI, 2012) on Mueller-Hinton agar after 24 hours incubation at 37 °C. Pathogens were tested with six different anbiotics: amoxicillin-clavulanic acid (AMC) (20 µg – 10 µg), trimethoprim-sulfamethoxazole (SXT) (1.25 µg – 23.5 µg), tetracycline (TE) (30 µg), enrofloxacin (ENR) (5 µg), lincomycin (MY) (2 µg), neomycin (N) (30 µg). The results were obtained by measuring the diameter of the growth inhibition zone around the antibiotic disc for isolated pathogens and recorded as susceptibility, intermediate and resistant. Pathogens recorded as intermediate were classified as resistant. Mathematical analysis was done by Microsoft Excel program. From all samples there were 29.6% infected and 5.3% of them infected by two pathogens. Coagulase negative staphylococci (CNS) were the most frequent pathogens isolated from the milk samples, and thus CNS presented 95.2% from all positive samples. The most effective antibiotics were trimethoprim-sulfamethoxazole and amoxicillin-clavulanic acid. The highest resistance was determined to neomycin. In conclusion, the most frequent pathogens in the milk of ewes were CNS indicating environmental sources of microorganisms causing subclinical mastitis and thus more effort should be focused on housing and milking hygiene. In a case of treatment, the trimethoprim-sulfamethoxazole and amoxicillin-clavulanic acid both seem to be more efficient.

Key words: *sheep, milk, pathogens, antibiotics resistance*

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The nutritive value of grape pomace silages with urea addition

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Supervisor: doc. Ing. Miroslav Juráček, PhD.

The objective of the research was to determine the influence of added urea on nutritive value of grape pomace silages. Fresh grape pomace (*Vitis vinifera* L.) of variety *Pinot Gris* with dry matter content 45% in laboratory conditions was ensilaged. Grape pomace was from University Experimental Farm Kolíňany. In the experiment were analysed two variants: first variant C (control-without additive) and second variant B with the addition of urea at rate of 4 $kg.t^{-1}$ on fresh grape pomace. Grape pomace from both variants (n = 3) was enclosed into the mini silo bags and hermetic sealed. After 6 weeks of storage in climatized laboratory conditions (t 22 ± 1°C). Average samples of grape pomace silages were taken for determination of nutrient's content in the Laboratory of quality and nutritional value of feed at the Department of Animal Nutrition at the Slovak University of Agriculture in Nitra. Application of urea influenced the nutritional quality of the grape pomace silages with statistically higher dry matter (5.50%), crude protein (22.75%) and statistically lower crude fiber content (13.94%) in comparison with control. The results confirmed, that the urea addition at rate 4 $kg.t^{-1}$ had positive effect on the protein value (PDIN by 22.79% and PDIE by 12.95%), but not influenced energy value of grape pomace silages. The net energy of gain value was statistically lower by 0.02 $MJ.kg^{-1}$ of dry matter after urea addition compared to control. From our results we can confirm that the addition of urea had a positive effect on nutritive value of grape pomace silages.

Keywords: *grape pomace silage; silo bags; nutritive value; urea*

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SECTION

Applied and molecular biology

Antioxidant activity of marigold (*Calendula officinalis*) extract on bovine spermatozoa

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Supervisor: prof. Ing. Norbert Lukáč, PhD.

The aim of our study was the evaluation of antioxidant effects of marigold (*Calendula officinalis*) extract and assess *in vitro* impact on selected quality parameters of bovine spermatozoa. Marigold is medicinal herb from the family *Asteraceae* native to sounthern Europe often used in pharmacology and medicine. A lot of positive properties include antioxidant, antibacterial, antiflammatory, antiviral, antifungal, antihelmintic and wound-healing activity of marigold. Flower extracts were subjected to high performance liquid chromatography (HPLC) which identified especially phenolic acids (rosmarinic and chlorogenic acids) and polyphenols (rutin, kaempferol, resveratrol, quercetin and apigenin). These substances are known for their antioxidant activity and positive effects against the oxidative stress. For our experiments, 10 samples of semen from sexually mature Holstein bulls were collected on a single day by using an artificial vagina, diluted in physiological saline solution and exposed to solutions with different concentration of marigold flower extract (75, 150 and 300 µg/mL). The samples with extract were cultivate at 37 °C in incubator. The selected quality parameters (motility, production of reactive oxygen species and level of lipid peroxidation) were analyzed after 0, 2 and 24 hours of *in vitro* culture. The motility evaluation was performed by using the computer-assisted sperm analysis (CASA) method. This method revealed that 75 and 150 µg/mL concentration of marigold flower extract had positive effect and increased motility of bovine spermatozoa compared to the control group ($P < 0.01$) in time 2 and 24 hours. This phenomenom was observed also in production of reactive oxygen species (ROS) and lipid peroxidation where was measured the malondialdehyde (MDA) production, which is the main product of lipid peroxidation. The concentration of 75 and 150 µg/mL had positive effects and decreased the levels of ROS and damages of membranes caused by lipid peroxidation compared to the control group ($P < 0.05$); ($P < 0.01$) in time 2 and 24 hours. Also the 300 µg/mL had the positivide effects but it was less significant than 75 and 150 µg/mL of extract . The data acquired from our study confirmed that 75 and 150 µg/mL of marigold flower extract had positive effect on motility of bovine spermatozoa, decreased the production of ROS and malondialdehyde in bovine spermatozoa. Based on results, the flower extract from marigold could be used for protection against oxidative stress in *in vitro* cultures.

Key words: *marigold, bull, motility, male gametes, antioxidant activity*

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The ability of streptomycetes to promote plant growth of arugula (*Eruca sativa*) and to suppress the activity of phytopathogenic fungi

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Supervisor: prof. Ing. Soňa Javoreková, PhD.

In agricultural environments, *Streptomyces* species are an important group of soil bacteria because of their ample capacity to produce PGP substances, secondary metabolites (such as antibiotics) and enzymes. Exhibit biocontrol action against a range of phytopathogens and promote plant growth thought production of phytohormones (IAA), siderophore and solubilize phosphate. The aim of our work was to determine the efect of actinomycetes on the growth of arugula and their ability to suppress the occurrence of a phytopathogenic fungus of the genus *Fusarium* sp. In this study 19 actinomycetes from Collection of microorganisms of the Department of Microbiology FBFS, SUA were tested. The isolates were characterized morphologically, biochemically and genetically and were subjected to a comprehensive *in vitro* screening for various plant growth promoting (PGP) traits and suppress the activity of phytopathogenic fungi. The most active indole acetic acid (IAA) producers were *Streptomyces flavovariabilis* KmiSK16A011, *Streptomyces puniceus* KmiSK16A004, *Streptomyces spectabilis* KmiICH17A097 and *Streptomyces olivochromogenes* KmiSK16A002. Only strains KmiICH17A097 and KmiSK16A011have been shown to solubilize phosphates and all tested strains have been shown to produce siderophores. The highest antifungal activity against phytopathogenic fungi *Fusarium graminearum* and *Fusarium culmorum* exhibited strains *Streptomyces albidoflavus* KmiICH17A098 and *Streptomyces sampsonii* KmiSK16A002. For *in vivo* screening, plant growth experiment was conducted by inoculating arugula (*Eruca sativa*) seeds with six the most active isolates and soil was inoculated with two phytopathogenic fungi *Fusarium* sp. Growth lasted 7 weeks and was carried out under controlled conditions (changes in temperature depending on the light phase – 16 hours, 28 ºC and dark phase – 8 hours, 22 ºC, constant humidity in both phases – 80%. Significant increase in root length was observed in variant with *Streptomyces olivochromogenes* KmiSK16A002 and maximum increases in plant fresh weight were recorded in variant with *Streptomyces flavovariabilis* KmiSK16A011. Over all the study revealed that these streptomycetes KmiSK16A011, KmiSK16A004 isolated from soil are good candidate to be developed as biofertilizer for promoting plant growth. The strain KmiICH17A098 is suitable for improving plant health because it suppressed the growth of phytopathogenic fungi *Fusarium* sp. on arugula.

Key words*: Streptomyces sp., PGP traits, phytopathogenic fungi, arugula*

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Antiradical activity of bioflavonoids

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Natural compounds are an inexhaustible source of drugs of various therapeutic effects. A study of the molecular mechanisms of the pathogenesis of a huge number of diseases of plants, animals, and humans has shown that all of them are associated with the activation or suppression of free radical processes. Therefore, the search and study of the regulators of such processes based on natural and synthetic raw materials remains relevant.

All plant compounds in relation to animal organisms have biological activity of an extremely wide spectrum, due to the diversity of their chemical structure, and are currently in the center of scientific attention. In connection with the foregoing, the search for antioxidants and the study of their inhibitory effect on the processes of free radical oxidation.

Based on this the antiradical activity (ARA) of the polyphenolic compounds - quercitin (Q), dihydroquercetin (DQ), rutin (R), gossypol (G), cynaroside (C) with respect to the stable free radical of DPPH (2,2-diphenyl-1-picrylhydrazyl) was studied.

To evaluate ARA in this work, we used a spectrophotometric method for measuring the kinetics of reduction of the molecules of the stable radical of 2,2-diphenyl-1-picrylhydrazyl (DPPH) by antioxidants. The test compounds were dissolved in alcohol at a concentration of 10^{-3} M.

When the test compounds are added to the alcohol solution of DPPH, the transition of free-radical molecules to a nonradical form occurs, while the intensely violet solution of DPPH becomes colorless.

To compare the ARA of the studied polyphenols, we selected a concentration of 50 μl for each compound from the prepared alcohol solution of the substance at a concentration of 10^{-3} M. Analyzing the results, we can conclude that the the optical density of the ethanol solution of DPPH decreases which indicates their antiradical ability.

From experimental data it follows that the studied compounds have a high ability to quench free radicals. To quantify the antiradical activity, we used the parameter t_{50} — the time required for the studied drugs to reduce the initial concentration of the radical by 50%, chemical reaction constant (k) and half inhibition concentration (IC_{50}).

| $K\,10^{-3}$, c^{-1} | | | | | IC_{50}, mcl | | | | | t_{50}, sec | | | | |
DQ	Q	R	G	C	DQ	Q	R	G	C	DQ	Q	R	G	C
1,2	5,3	4,8	5,0	1,1	14,3	7,2	9,7	8,4	15,2	105	9,6	28	24	152

Thus, the studied compounds are characterized by strong ability of components to react with free radicals.

Key words: *polyphenols, antiradical activity, free radicals*

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Determination of evaluation of cyclodextrins efficiency as solubilizers for cynaroside

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Supervisor: prof. Takhir Aripov, academician

The success of modern therapy is due not only to the creation of new highly effective drugs but also to the optimization of existing and the development of fundamentally new dosage forms. This work was dedicated to exploring the possibility of increasing the water solubility of flavone glycoside – cynaroside. As cynaroside proved to be an effective hypoazotemic agent and has been doing developed in tablet forms in Uzbekistan, there was a need to continue the development of this drug in other accessible forms.

In this work, we studied the effect of native γ-cyclodextrin (γ-CD) and modified hydroxypropyl-β-cyclodextrin (HP-β-CD) on increasing the solubility of cinaroside, determining the physicochemical parameters of the process, and establishing the possibility of using these cyclodextrins as solubilizers for cinaroside.

We used the dimensionless UCD value (cyclodextrin utility number) - an assessment of the efficiency of the CD (minimal value for effective complexation $\geq$ 1) - and calculated hypothetical complexation stability constant (K) while assuming 1:1 ratio (cynaroside:CD). The theoretical K value for γ-CD as a solubilizer was 24,021.08 M^{-1} (the experimental was 54,741.06 M^{-1}), for HP-β-CD was 2,154.321 M^{-1} (the experimental value was 21,763.56 M^{-1}). The calculated free Gibbs energy (ΔG) and efficiency constant (CE) were: ΔG (cynaroside:γ-CD) = -27,049.7; ΔG (cynaroside; HP-β-CD) = -24,762.8; CE (cynaroside:γ-CD) = 0.054741 indicates that approximately one of every 19 γ-CD molecules forms a complex with cynaroside; CE (cynaroside; HP-β-CD) = 0.021764 indicates that approximately one of every 46 HP-β-CD molecules forms a complex with cynaroside. Also, according to the literature negative ΔG values indicate favorable conditions, stability constants (K) values around 100 – 5,000 seem to be suitable for practical applications.

Thus, we have found that both cyclodextrins could be used for improving cynaroside solubility while HP-β-CD seems to be more appropriate. Our results also reflect the wide usage of modified β-CDs in pharmacology, cosmetics, etc. and support further development of cynaroside in new dosage forms.

Key words: *solubility, free energy, complexation efficiency, complexation stability*

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Isolation and characterization of *Bacillus* strains and their effect on improving growth of maize (*Zea mays* L.)

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Bacteria living in the soil as free organisms or as endophytes, that promote plant growth and protect plants from disease are called Plant Growth Promoting Bacteria (PGPB). *Bacillus* species are the predominant PGPB of many crops. Abiotic and biotic stress factors that have harmful effects on crops are alleviated by the physiological changes induced by *Bacillus*. The aim of this study was to (1) identify *Bacillus* species isolated from maize roots, (2) to determine biochemical properties that promote plant growth and the ability to protect plant against pathogens and (3) to monitor changes in plant growth after application of *Bacillus* strains to the maize seeds under *in vivo* conditions. We identified three strains 231K, 239K and 246K, which were isolated from maize (*Zea mays* L.) roots. For rapid identification was used analysis of proteins by MALDI-TOF mass spectrometry. *Bacillus* strains were tested for (1) production of indole-3-acetic acid (IAA), (2) production of siderophores using CAS medium, (3) ability to dissolve phosphates and (4) ability to inhibit the growth of phytopatogenic fungi *Fusarium* sp. Maize seeds were soaked in a bacterial suspension of *Bacillus* strains with concentration of 0.5 Mac Farland (1.5×10^8) and applied to the organically poor soil. Plant growth was performed under controlled conditions in a cultivation chamber for 4 weeks. The conditions were as follows: light phase for 16 hours with temperature 28 °C and dark phase for 8 hours with temperature 22 °C and 80% humidity in both phases. An untreated plant was used as a control. *Pseudomonas simiae* strain CCUG 50988 (Netherlands) was used as a positive control. We observed the length and weight of the root system. Bacterial strains were identified as *Bacillus megaterium* (231K), *Bacillus flexus* (239K) and *Bacillus subtilis* (246K). All strains produced phytohormone indole-3-acetic acid (IAA), showed positive siderophore production and achieved the middle phosphate solubilization index ($2.00 \leq SI < 4.00$). All *Bacillus* strains inhibited the growth of *Fusarium* sp. to more than 50%. All three *Bacillus* strains promoted the length and weight of the maize root system under *in vivo* conditions. The length of the root system increased by more than 9.9 cm and the weight increased by more than 5.37 g on average according to the control. The length of the root system increased by more than 6.62 cm and weight increased by more than 1.05 g on average according to the positive control (*Pseudomonas simiae*). The effect of three *Bacillus* strains on maize seeds *in vivo* conditions was positive due to an increase in the length and weight of the root system compared to the control and positive control.

Key words*: plant growth promoting bacteria, Bacillus, biochemical features, in vivo experiment, maize*

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Actions modified sulphated cellulose for thrombin time

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Supervisor: prof. Kabil Nasirov

The experiments investigated the effects of modified sulphated cellulose SC-GSC-66 on thrombin time (TT). In laboratory conditions, the final stage of the coagulation cascade is reproduced by adding ready thrombin to the plasma, and changes in the structure or concentration of fibrinogen are reflected in an increase or decrease in thrombin time. Experiments were performed in the presence of 50 μg/ml. SC-GSC-66 was preincubated with fibrinogen, then thrombin was added, while the preparation at a concentration of 50 μg/ml did not cause a change in thrombin time relative to the control. But, if the SC-GSC-66 preincubated in plasma with thrombin, calcium, and then fibrinogen is added to it, these drugs at a concentration of 50 μg/ml caused an increase in thrombin time. The results show that SC- GSC- 66 does not affect the activity of fibrinogen, but mainly act on the earlier stages of the conversion of prothrombin to thrombin. In these experiments, it was found that the SC- GSC-66 prolonged the thrombin time to varying degrees. When studying the effect of SC- GSC-66, depending on the dose, it was found that SC-GSC-66 at a concentration (5 – 50 μM) at normal (18 – 25 sec) lengthened TT to 140-160 sec. Among them, the most inhibitory concentration of 50 mg/ml extends the thrombin time to 160 seconds. Thus, it can be assumed that SC-GSC-66 at concentrations (5 – 50 μM) lengthened the time of thrombus formation, relative to the control, leading to a weakening of the formation of a fibrin clot, which may indicate inhibition of the activity of one of the factors IXa, Xa, XIIa and antithrombin III.

Key words: *hemostasis, anticoagulant, platelet aggregation, sulphated polysaccharide*

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Anticoagulant activity of sulphated polysaccharides

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The aim of the study was a comparative analysis of the effect of various modified sulfated polysaccharides (MSPs) on individual links of coagulation hemostasis, anticoagulant activity of rat blood plasma *in vitro*. The experiments showed that the anticoagulant activity under the action of polysaccharides increases in a dose-dependent manner. The anticoagulant activity of some MSPs is associated with the ability to increase the rate of inhibition of serine proteases of the blood coagulation system, thrombin (factor IIa), and factors Xa, IXa, XIa, XIIa, mainly activating the plasma inhibitor-antithrombin. In *in vitro* experiments on rat blood plasma, it has been shown that MSPs exhibit anticoagulant activity similar to heparin. Compounds had a different effect on coagulation hemostasis of blood plasma. It was shown that in concentrations from (5 – 50 μM) modified polysaccharides lengthened the clotting time of rabbit blood plasma, increase the time of fibrin clot formation in vitro in standard coagulological tests, in tests for prothrombin time, APTT, thrombin time. *In vitro*, at different concentrations of compounds from 5 to 50 μM, their anticoagulant activity was detected, which increased by 20 – 25% compared with the control and corresponded to the anticoagulant action of the equivalent dose of heparin. *In vitro*, we also showed that the compounds inhibit the activity of thrombin by 18 – 20%. Thus, the data obtained suggest the need for a deep study of sulfated polysaccharides as blood anticoagulants. The study of the connection structure - the activity of cellulose sulphate will make it possible to get the most active polysaccharides directed.

Key words: *hemostasis, anticoagulant, platelet aggregation, sulfated polysaccharide*

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Antinuclear autoantibodies: our friends or enemies?

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The recognition of the antigen by the specific antibody is the key reaction in the immunity of gnathostomate vertebrates. Although the beneficial role of this phenomenon in the elimination of infections, it may also occur in the processes which damage cells and tissues of the own organism. The destructive, chronic inflammation is one of the most important and still not resolved problem in the contemporary medicine. In the great majority of autoimmunological disorders, the antinuclear autoantibodies are detected, but their role in the etiology and pathophysiology of autoimmunological diseases is not explained. We still do not know if they are a cause or only a molecular symptom of the autoimmunity. Furthermore, they are detected in several diseases, which have not an autoimmunological background, i.e.: microbiological infections, tumors, drug–derived complications, parasitological invasions, etc. We analyzed serological examinations of 61 patients (33 males and 28 females) without any manifestations of autoimmunological disorders, but with oncological, endocrinological or infective diagnoses. The antinuclear autoantibodies were detected using the intermediate immunofluorescence assay (IIF) and the double immunodiffusion assay (DID) in the plasma of the blood. In 92% of examined individuals (91% of males and 93% of females) we found the presence of non-specific antinuclear autoantibodies (granular lighting signal in the dilution 1:160 or more). 19 cases of them were connected with the male hypogonadism disorders, 11 with the female hyperestrogenism, 11 with the severe common acne, 6 with dermatofibroma non–malignant proliferative disorders, 5 with the female hyperprolactinemia disorders and 4 with the skin cancer. The common occurrence of non-specific antinuclear autoantibodies in the cases of non-autoimmunological diseases leads to the conclusion that this serological test is not a good diagnostic parameter for autoimmunological diseases, which should be always verified using a specific Western Blot assay. We also suggest that the antinuclear autoantibodies may be involved in the reaction of the organism against cancer cells and certain kinds of microorganisms. It is also possible that many hormonal disturbances stimulate the production of autoreactive antibodies. Finally, we discuss the modern opportunities in the modulation of the autoagressive immunological response, i.e.: immunosupression, immunostimulation, hormone therapy, biological medicines. It is evident that the physiological and pathophysiological role of the antinuclear autoantibodies requires further, extended and interdisciplinary explanation.

Key words: *antibody, autoimmunity, immunofluorescence, immunomodulation, inflammation, lymphocytes*

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Calgonit for industrial synthesis

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Calgonit, Calgon, sodium hexametaphosphate or simply polyphosphate (polyP) are synonyms for worldwide used chemical water softener. However, in addition to cleaning, polyphosphate also fulfils other irreplaceable tasks. In microbial cells, polyP granules present easy and rich source of energy and are also used in production of nucleotide triphosphate or phosphate. The metabolism of polyphosphate formation/degradation is controlled by a group of enzymes called polyphosphate kinases (PPKs), which can be categorized into three groups (PPK1, PPK2 and PPK3) according to their functionality. Here we present characterization and comparison of two homologs of PPK2 in the form of active inclusion bodies. In general, a group of PPK2 homologs degrades polyP and phosphorylates nucleoside diphosphates. Selected enzymes, PPK2 *Coraliomargarita akajimensis* (*cor*PPK2) and PPK2 *Magnetococcus marinus* (*mag*PPK2) were fused with cellulose binding domain (CBD) from *Clostridium cellulovorans* and expressed in *Escherichia coli*. After isolation and purification of IBs, we determined optimal temperature conditions, pH and magnesium concentration. We also compared the preference of enzymes in different nucleosides diphosphates phosphorylation. Capillary electrophoresis was used to analyze all samples. Based on the obtained results, we assumed and subsequently confirmed which PPK2 is more suitable for our final experiments: synthesis of CMP-sialic acid (CMP-sia) and synthesis of UDP-glucose (UDP-glu). Both, CMP-sia and UDP-glu have irreplaceable role in saccharide metabolism. CMP-sia acid is substrate for sialyltransferases and is used for the enzymatic sialylation of glycans. UDP-glu present activated form of glucose and is used as substrate for enzymes called glucosyltransferases.

Key words*: calgonit, polyphosphate kinases, inclusion bodies, CMP-sialic acid, UDP-glucose*

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Morphological characteristics as a key attribute for a successful determination of selected *Cotoneaster* species – preliminary results

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The classification of taxa included in *Cotoneaster* genus is relatively problematic, because morphological features, such as colour of fruit and bark or shape and size of leaves, are not always reliable attributes. The western part of Carpathian Mountains includes 4 different *Cotoneaster* species – *Cotoneaster integerrimus* Medik., *Cotoneaster matrensis* Domokos, *Cotoneaster melanocarpus* (Bunge) Fisch. et C. A. Mey and *Cotoneaster tomentosus*. The number of pyrenes in fruit and the number of fruit in infructescence are the most important morphological features for determination of these *Cotoneaster* species. Results obtained from the last study by using a flow cytometry method revealed no significant difference in genome size of tetraploid *Cotoneaster* species (*C. integerrimus* and *C. melanocarpus* agg. including *C. matrensis*). In this study, we decided to use previously mentioned morphological features for determination of closely related tetraploid *Cotoneaster* species and to prove whether it is a reliable characteristic for identification of these species. Samples were collected from various localities in the Western Carpathians (Slovakia and Hungary). The collection of samples, designed for counting of pyrenes in fruit, included 1987 fruit of > 80 individuals collected from 32 localities. The collection of samples, designed for counting of fruit in infructescence, included 450 infructescences of 46 individuals collected from 17 localities. Number of pyrenes in fruit ranged from 1 to 5. Statistical analysis revealed a significant difference in the mean number of pyrenes in fruit between all three *Cotoneaster* species – *C. integerrimus* (2.91), *C. melanocarpus* agg. (2.46; including *C. matrensis*) and *C. tomentosus* (4.18). Number of fruit in infructescence ranged from 1 to 4. Statistical analysis also revealed a significant difference in the mean number of fruit in infructescence between *C. integerrimus* (1.20) and *C. melanocarpus* agg. (1.56; including *C. matrensis*). Results from this study proved, that even in the era of modern cytological and molecular methods, classic morphology methods are still relevant in the field of botany.

Key words: *classification, Cotoneaster, pyrenes, infructescence, morphology*

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Relaxant effect of the F–37, an isoquinoline alkaloid on isolated rat aortic smooth muscle

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Isoquinoline alkaloids constitute one of the largest groups of plant – derived compounds with a wide range of pharmacological activities are a promising candidate for developing new cardiovascular drugs. The aim of the present study was to determine the relaxant effect of 1-(4′-Chlorophenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (F – 37), an isoquinoline alkaloid in the rat isolated aorta smooth muscle cells (SMC). The experiments were performed in preparations, which are ~3 – 4 mm wide rings isolated from aortic albino rats (~200 – 250 g) and placed in a special chamber (5 mL) was perfused with solution Krebs – Henseleit following composition (mmol/L): $NaCl$ – 118.6; KCl – 4.8; $CaCl_2$ – 2.5; $MgSO_4$ – 1.2; KH_2PO_4 – 1.2; $NaHCO_3$ – 20, glucose – 10 (pH = 7.4). During the experiments, while working with experimental animals, International principles of The Council for International Organizations of Medical Sciences and the rules of human attitudes towards animals were completely followed (2012). Force contraction was recorded using an FT.03 (Grass – Telefactor, USA). In the experiments, pretreatment with F – 37 (1 – 100 µmol/L), attenuated the relaxant effect of the isolated rat aorta from $91.1 \pm 3.6\%$ ($IC_{50} = 15.7$ µmol/L) in comparison with the control group ($p < 0.05$; $n = 3 – 4$). Studies have shown that in incubation to inhibit potential-dependent Ca^{2+}– channel – verapamil ($IC_{50} = 0.1$ µmol/L) the relaxant effect of F – 37 ($IC_{50} = 15.7$ µmol/L) decreases to $41.6 \pm 3.9\%$ of control values. It is known that KCl – induced reduction of SMC aorta is associated with activation of potential – dependent Ca^{2+}– channels of plasma membranes of SMC. At the same time, the increase $[K^+]_{out}$ changes the membrane potential and causes depolarization, due to it activates potential – dependent Ca^{2+}– channels, which leads to an increase $[Ca^{2+}]_{in}$, which in turn causes a reduction of SMC. In preliminary experiments, it was shown that F–37 (100 µmol/L) on the back of the aorta contraction induced by phenylephrine (1 µmol/L) and in the presence of verapamil, a specific blocker of potential-dependent Ca^{2+}– channels (0.1 mmol/L), caused a suppression of power cuts SMC rat aorta to $95.9 \pm 3.8\%$ ($IC_{50} = 25.5$ µmol/L), relative to a control ($n = 3 – 4$, $p < 0.05$). It was established that the relaxant effectively of F – 37, an isoquinoline alkaloid in conditions of KCl – and phenylephrine – induced contraction was related to the inhibition of the influx of Ca^{2+} ions through the voltage – dependent and receptor-operated Ca^{2+}– channels SMC.

Key words: *isolated rat aorta, isoquinoline alkaloid, relaxant effect, Ca^{2+}–channels*

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SECTION

Nutrition

The impact of addition of different tea powders on the biological activity of white chocolate

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Supervisor: prof. Ing. Miroslava Kačániová, PhD.

Dark chocolate is usually considered as the excellent source of bioactive compounds, especially polyphenols. On the contrary, white chocolates are generally deficient in polyphenolic compounds. However, since white chocolate is one of the most favoured types of chocolate not only among children, but also among adults, and only perceived as a sweet treat, it was evaluated for the possible changes after the addition of tea powders. For that reason, this work focused on the assessment of the potential positive effects of teas added to the white chocolate and the impact of such enrichment on the subsequent biological value and antioxidant activity. In order to compare the selected parameters, an array of rapid and reliable, widely used spectrophotometric methods for evaluating the biological activity were applied. For assessing antioxidant activity it is advised to use at least two different methods to provide more reliable data. For this reason, we used methods using DPPH and ABTS radicals and even reducing power method to evaluate this parameter. The acquired results proved that plain white chocolate itself is only a poor source of biologically active compounds and also antioxidant activity measured by different methods was found low. Nevertheless, the addition of various types of teas to white chocolate significantly enhanced the number of total polyphenols, flavonoids, and phenolic acids. Addition of green teas increased the total content of polyphenols more than four times in comparison to the control. Such enrichment also influenced the antioxidant capacity of the studied samples positively. High positive correlation ($p \leq 0,05$) was found between ABTS and DPPH assay results (r = 0.9882) and also between ABTS and phenolic acids content (r = 0.9946). The best results were obtained with the addition of matcha green tea.

Key words: *antioxidant activity, enrichment, polyphenols, tea, white chocolate*

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Effect of ketogene diet on changes of anthropometric parameters of selected group of women

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Ketogenic diet is a nutritional regime characterized by very low carbohydrate levels (less than 20 g per day, or 5% of total daily energy intake) and a relative increased protein and high fat content. Low carbohydrate levels forces the body to use fat as the primary source of energy. Anthropometric methods are among the simplest, cheapest and non-invasive procedures for measuring body dimensions. Body mass index is most often used (BMI), waist-to-hip (WHR), skeletal muscular mass (SMM), body fat (BFM) and visceral fat area (VFA). The aim of the study was to investigate the effect of ketogenic diet on weight loss monitoring of selected parameters - BMI, WHR, SMM, BFM and VFA. The study, which took three weeks, was attended by 11 women. Anthropometric measurements were performed by InBody 230 (Biospace Co. Ltd., Seoul, Korea Republic), working on the basis of bioelectric impedance. To process the results, we used Lookin'Body 3.0 software. Data from anthropometric measurements are statistically and graphically evaluated in Microsoft Office Excel 2010 (Los Angeles, California, USA). Statistical analyzes were performed using STATISTICA Cz version 10. The average weight reduction of respondents was 3.1 kg (3.21% of the original body weight). The average fat loss was 3.0 kg (10.5% of the original BFM). The decrease of the mean BMI correlates with a decrease in body weight. BMI decreased 31.2 kg/m^2 to 30.2 kg/m^2. WHR index is not changed. There was a slight increase in skeletal muscle by 0.8 kg. Weight loss is caused by a reduction in body fat. The results indicate the effectiveness of a ketogenic diet, at least in the short to medium term and can be an effective tool to reduce fat mass and visceral fat issue.

Key words: *ketogenic diet, body composition, anthropometry, bioimpedance, visceral fat area*

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Determination of silymarin complex using high-performance liquid chromatography and its application in cereal products

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Supervisor: doc. Ing. Miroslav Habán, PhD.

Milk thistle (*Silybum marianum*) extract has been employed as traditional herbal remedy for almost 2000 years and currently it is one of most commonly used botanical supplement in the world. The ease of availability, comparative low cost and safety profile provide advantages for its usage as a potent drug with immense clinical benefits. Most of documented data with Silybum marianum are connected with liver disorders; however, its several health-promoting actions on a wide variety of other diseases have been recently described. Indeed, silymarin acts as an anticancer, anti-inflammatory, immunomodulatory, neuroprotective and lactogenic agent. The main active component of milk thistle known as silymarin is extracted from the seeds of the plant. Silymarin consists of a mixture of flavonolignans and flavonoid taxifolin. The major flavonolignans present in most extracts are silybin A, silybin B, isosilybin A and isosilybin B, silydianin, silychristin and isosilychristin. A high-performance liquid chromatographic method was developed for the determination of the silymarin flavonolignans. The method will be used and validated in our study for measurement of these valuable biological active compounds in the achenes of milk thistle varieties growing in different agroecological conditions. In the next step, nanotechnological methods for silymarin encapsulation in order to improve its bioavailability and provide its prolonged-release characteristics at the site of absorption will be applicated. Finally, encapsulated silymarin will be incorporated into bread formulation and the changes in rheological properties of wheat flour dough with addition of silymarin, as well as volume, textural and organoleptic properties of silymarin-enriched bread will be evaluated.

Key words: *Silybum marianum, silymarin, flavonolignans, high-performance liquid chromatographic, encapsulation, bakery products*

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Starch and glycemic carbohydrate content in non-traditional cereals

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Cereals are major sources of carbohydrates. Nowadays, there is a renewed interest in using non-traditional wheat species to make baked products. Spelt, emmer and einkorn have been considered as particularly interesting from a nutritional point of view. They can be ideally suited for organic farming. The present work was aimed at the evaluation of nutritional quality of carbohydrates, digestible/ glycemic carbohydrate content in bread prepared from whole grain flour and flour and to compare starch content in *Triticum* species. Field experiments were carried out at Dolná Malanta (experimental station) from 2015 to 2017. Laboratory samples were analysed in the Laboratory of Department of sustainable agriculture and herbology of the SAU in Nitra and in Research Institute of Crop Production in Prague. Experimental materials were *Triticum aestivum L., Triticum monococcum L., Triticum dicoccon* and *Triticum spelta L.* For the evaluation of data was used MS Office Excel and statistical software STATISTICA version 10.0. Available carbohydrates (ACH) were analysed in bread according to AACC 32-05.01, 32-07.01, 32-21.01 methods, a kit Megazyme K-ACHDF 06/ 18 was used. This method involves digesting the food with suitable enzymes and determination of glucose (D-Glu) and fructose (D-Fru). The starch was analyzed by Ewers polarimetric method following the ČSN EN ISO 10520. The baking formula was: flour (600 g, 14% mb), compressed yeast (9.46 g), salt (7.89 g), sugar (9.78 g). Doughs were mixed, left to rise for 30 min in fermentation chamber. The dough was punched, molded, put into a baking pan and left for fermentation for further 70 min. After adequate proofing, doughs were baked for 20 min at 225 °C. There was a statistically significant effect of T. species on the content of ACH and D-Glu. The lowest value of ACH was determined for T. *monococcum L.* (59.8 g) and the highest value for *T. dicoccon* (68.3 g). The content of   D-Glu follow the findings of ACH. There were differences in the content of ACH also between the type of flour. The average amount of ACH in whole-grain flour was 60.1 g and in flour was 68.3 g. There was not statistically significant effect of T. species on the content of D-Fru. The average amount for D-Fru was 0.05 g in all species. T. species had a statistically significant effect on the starch content too. The lowest value of starch was determined for T. *monococcum L.* (61.2 g) and the highest value for *T. dicoccon* (70.6 g). There were differences in the content of starch also between the years. In 2016 was the highest value for starch (68 g).

Key words*: non-traditonal cereals, starch, glycemic carbohydrates*

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SECTION

Multifunctional agriculture, environment, landscape architecture and rural development

Scientific Conference of PhD. Students of FAFR, FBFS and FHLE SUA in Nitra – Proceedings of Abstracts
SECTION **Multifunctional agriculture, environment, landscape architecture
and rural development**

Assessment methodology of plant species composition suitability for diverse green space categories in the Slovak countryside

Denis BECHERA

Supervisor: Assoc. Prof. Dr. Gabriel Kuczman

Trees and herbs are a natural part of the Slovak rural landscape. They significantly influenced the landscape, its overall character, the urban structure of the countryside and its green infrastructure. Their significance was not only aesthetic, but also of great importance as elements contributing to the cultural expressions of society and the degree of maturity of the creators of the cultural environment. Trees and herbs in the composition, planting and architectural design supported the identity of the place and the "Genius loci" not only of extraordinary places, but also of commonplaces, villages and spaces in the countrysides. They distinguished the individual regions of Slovakia and thus contributed to the uniqueness of the places. The rural landscape is becoming increasingly productive and commercial. The identity, character and expression of the Slovak countryside are often lost. It is becoming a part of the urban agglomeration, which relates to the functional use of the areas and the aesthetic demands of the inhabitants and the management of the towns on the environment. This trend also has adverse effects on the ecosystem, the environment, biodiversity, the overall development and direction of the rural landscape. Currently, modern elements with mostly highly aesthetic properties overlap with the original ones.

The aim of the research task was to develop a framework methodology for assessing the suitability of species composition of plants for functional categories of greenery in Slovak rural conditions. This methodology will be implemented in specific areas public areas of selected rural settlements in the next phases of doctoral research.

The presented methodology is one of the first partial results of the doctoral research. It was created on the basis of empirical experience from previous scientific research activities and identified requirements of landscape, architectural practice. The doctoral research will apply the assessment methodology in model areas, focusing on the assortment of plants and compositional solution of public spaces. The results will be a better knowledge of the functional and spatial use of plants in the Slovak countryside under conditions of changing climate.

Key words: *countryside, rural greenery, greenery in rural areas*

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Comparison of two different potential bioavailable forms of mercury in Technosol developed on the heap after mercury mining activity

Nikola BENKOVÁ, Pavol MIDULA, Oliver WICHE

Supervisor: doc. Ing. Radoslava Kanianska, CSc.

Mining activities are important contributors to environmental contamination. The impact of mining on the environment is often manifested even after its termination. Negative consequences can be observed in all environmental elements, including soil and biota. In our research, we measured the total content of mercury in the soil (Technosol) as well as the content of potentially bioavailable forms of mercury obtained by extraction with ammonium acetate of pH 7 (neutral environment simulation) and pH 5 (acid environment simulation) at the Veľká Studňa dump-field (Malachov, Slovakia). We collected 12 soil samples from A horizons which were subsequently subjected to processing and advance preparation for analysis by ICP-MS (inductively coupled plasma mass spectrometry). The mean content of total Hg in soil was 435.5 ± 283.2 mg.kg^{-1} dry mass (range $140.3 – 910.8$ mg.kg^{-1}). Such values indicate an important increased concentration of mercury in the collected soil samples because exceed many times the limit values set by the valid legislation of the Slovak Republic. The mean content of Hg after the neutral extraction was 1.3 ± 2.6 µg.kg^{-1} dry mass (range $0 – 8.0$ µg.kg^{-1}). The mean content of Hg after the acid extraction was 11.9 ± 9.9 µg.kg^{-1} dry mass (range $0.8 – 37.5$ µg.kg^{-1}). The fraction extracted with ammonium acetate of pH 7 brought lower contribution to the total Hg content (0.0003%) compared to the fraction extracted with ammonium acetate of pH 5 (0.0027%). Despite the low content and percentage of both fractions on total Hg content in soil they are relatively mobile that can easily enter plants and accumulate in living organisms. Mercury is known to be one of the most toxic elements and even low concentrations may cause serious threat to environment and health problems.

Key words*: mercury, potential bioavailability, soil contamination, ammonium acetate, Malachov*

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Re-designing public space - analysis, data and quick solutions

Miroslav ČIBIK, Roberta ŠTĚPÁNKOVÁ

Supervisor: doc. Ing. arch. Roberta Štěpánková, PhD., Ing. Attila Tóth, PhD.

The article discusses how the design of high-quality public space can be created with the analyses, data collection and quick solutions and how the space function can be restored. As an example, we present two square suggestions where the function changes from communication to residential. Our landscape-architectural designs solve the space comprehensively and bring to the place life, atmosphere and attractiveness. The first example is Svätopluk Square in Nitra. In this area we consider the fountain as the most attractive element. Through it, as well as the imaginary centre of the theatre, which we perceive as a fundamental, representative element is a circle that defines two main interventions in the square. The third intervention is formed by the closer surroundings of the fountain itself. Sculptures appear on the axes that we lead through the centre of the theatre, confirming the above-mentioned landmark and its view. The second example is the Majzon Square in Nové Zámky. We focused through detailed analyses of the current state on the overall view of the area. The main element of the whole design is clearly the dominant feature of the space in the form of a water reservoir of organic shape located on the axes of the two main pedestrian routes through the territory. The plan view of the water element represents the history of Nové Zámky, where the gutter covered with hardened glass defines the historic buildings of the original fortifications, the central water surface represents the original meander of the river and again the organic footprint. The gutter is lined with a light element representing the castle tower of the original fortification. We have combined water retention measures with a design that bears a historical legacy, and therefore future users of public space will perceive the water in the area much more intensively. We did not omit any greenery in the design, when we added new domestic species of deciduous trees and perennial. The beds are lined with attractive wooden seating, which also communicates better and provides access to large areas of greenery for people. They will be irrigated by rainwater from the surrounding flats, as well as by excess water circulated from underground reservoirs. This circulation appears on the surface as a design water feature. Our idea was to create a high quality public space that connects blue and green infrastructure into a sustainable multifunctional element in the city centre, which in our view is a huge benefit for the future and can help the city considerably.

Key words: *public space, landscape architecture, re-functioning space, Nitra, Nové Zámky*

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Preferences of schoolchildren in the everyday use of schoolgrounds

Eszter JÁKLI

Supervisor: Zsombor Boromisza PhD.

Understanding and adjusting to the preferences of potential users and participatory planning itself are becoming more and more common throughout the design process in landscape architecture. Regarding schoolgrounds, there is a particular group that we need to work with: children; and there is a particular topic that this research is focusing on: environmental awareness and attitudes. The study aims to discover how children use the open spaces of schoolgrounds, and whether they prefer more natural environments if they have the possibility or not. The hypothesis is that as natural biophilia is still present in children, when they are given the chance to play in (semi)natural or nature-like environments, they tend to choose these rather than man-made environments. 8- and 10-year-old schoolchildren in eight classes of four primary schools were surveyed. Besides basic demographic questions and questions about their living environments, basic knowledge of plants and animals, environmental attitudes were measured by simple questions, focusing on their use of the outdoors. In the second part of the survey, children were asked to answer shortly some questions - either by words or by drawing – about their schoolgrounds: which is their favourite activity and favourite area and what they would like to add to the schoolgrounds. The results of the survey were compared to the physical layout of the schoolgrounds in order to understand whether the landscape design and the physical characteristics influence the choices of the children. The study concludes that natural play possibilities and semi-natural or nature-like areas on schoolgrounds are appreciated by children, however sport – especially football – and playing are more common and are mentioned more.

Key words*: schoolgrounds, landscape architecture, user preferences, children*

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Soil organic matter vs. soil structure after biochar application

Martin JURIGA

Supervisor: doc. Ing. Vladimír Šimanský, PhD.

The biochar application has the potential to increase the content of soil organic matter (carbon sequestration) and to enhance the state of soil structure. Mainly due to increasing atmospheric CO_2 concentration and global warming, the need to increase the retention of organic matter in the soil is rising. The study was carried out from 2017 to 2018 with the aims: (1.) to compare the effects of different biochar rates (0, 10, 20 t ha^{-1}) on the contents of soil organic carbon (C_{org}) and labile carbon (C_L), the size fractions of water stable aggregates (WSA $> 5, 5 - 3, 3 - 2, 2 - 1, 1 - 0.5, 0.5 - 0.25$ and < 0.25 mm), the contents of C_{org} and C_L in WSA, (2.) to determine of interrelationships between C_{org}, C_L contents and WSA size fractions, (3.) to evaluate the impact of changes in the C_{org} and C_L contents in WSA on the C_{org} and C_L contents in the soil from 2017 to 2018 and (4.) to assess the carbon sequestration capacity in the soil as well as in the WSA. In 2014, the field experiment with biochar was established on experimental site of SUA in Nitra region (Dolná Malanta) on a silty loam Halpic Luvisol. The soil samples were taken at monthly intervals during the vegetation seasons of corn (2017) and spring barley (2018).

The results showed that the biochar application was more effective at the rate of 20 t ha^{-1} compared to 10 t ha^{-1}. In B20 treatment, the content of WSA in larger size fractions was significantly increased: > 5 mm (+ 68%), $5 - 3$ mm (+ 64%), $3 - 2$ mm (+ 48%), $2 - 1$ mm (+ 43%) and the content of WSA in smaller fractions were reduced: $1 - 0.5$ mm (- 21%) and < 0.25 mm (- 41%) compared to control (B0). In addition, the C_{org} content in the soil, C_{org} in all WSA fractions and C_L in WSA 2-1 mm and 0.5-0.25 mm also increased. The C_L content in the soil contributed to WSA 5-3 mm (r = 0.891***) and $2 - 1$ mm (r = 0.755**) formation. At the same time, C_{org} in WSA $3 - 2$ and $2 - 1$ mm were in negative correlations with WSA > 5 mm (r = - 0.684*) and (r = - 0.801**) and in positive relationships with WSA $0.5 - 0.25$ mm (r = 0,670*) and (r = 0,656*). From 2017 to 2018, in all treatments, the C_{org} content in each WSA fraction negatively affected the change in the C_{org} content in the soil. While in B20, the C_L content in the most of WSA fractions contributed favourably to the change of soil C_L content, in B0 the C_L content in the most of WSA fractions contributed negatively. Furthermore, the application of a higher biochar rate had also a positive effect on the increase of soil carbon sequestration capacity (CSC) (+ 21%). The CSC values in WSA fractions varied within the treatments. In B20 treatment, CSC value increased in WSA > 5 mm (+ 16%), $5 - 3$ mm (+ 12%), $3 - 2$ mm (+ 4%), $1 - 0.5$ mm (+ 6%), $0.5 - 0.25$ mm (+ 7%) and < 0.25 (+ 7%) and decreased in WSA $2 - 1$ (- 13%) compared to B0.

Key words: *biochar, carbon sequestration, soil aggregates, soil organic matter*

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Natural conditions and their influence on dendrometic changes of *Juglans regia* L.

Martina KOVÁČOVÁ, Viliam BÁREK, Vladimír KIŠŠ

Supervisor: doc. Ing. Viliam Bárek, PhD.

The global climate change is currently at the Slovakia manifested as increase of the average annual temperature of 1.73 °C. Decrease of the total amount of atmospheric annual precipitation of 0.5%, while on the south part of the territory was decrease by more than 10%. On the north and northeast part was measured local increase of the precipitation to 3%. The result of these changes is drought, which causes changes in the ecosystems, biodiversity and also expected change of the vegetation levels and zones boundaries (MŽP SR, SAŽP, 2018). On the base of climate change it is necessary to optimize amount of irrigation dose and its timing according to vegetation needs.

The aim of the work was monitoring dendrometrical changes of vegetation according to natural conditions and thus to define time water deficit in plants. For research was chosen subject of Iranian walnut (*Juglans regia L.*) in orchard situated in Nové Zámky. To measure diameters of branches we used sensors Diameter Dendrometer Small (DD – S) and as datalogger was used DL – 18 from Ecomatik. Based on data from meteorological station placed in orchard we got overview about real conditions, which had impact on our monitored subjects. In 2018 was the range of the temperature between 8.5 °C and 31.5 °C and the highest difference was measured between June 21 and June 22. Rainfall wasn´t evenly distributed. The biggest amount of the crops was in the begin of the vegetation season, in the first half of July with maximum height 15.20 mm. Temperature course in 2019 was more evenly and also changes of temperature wasn´t that critical in short time. Precipitation conditions were more optimal considering to their division in second half of June. The result of these measurements is finding, that despite of higher precipitation values in earlier vegetation stage in 2018 the growth was slower in comparison with 2019, when precipitation conditions in earlier vegetation stage was worse. In first half of June 2019 was occurrence of precipitation lower but more evenly so the irrigation of the plant was more optimal. In 2018 the largest expansion of plant biomass was measured on June 21, between 9 pm and 10 pm. The increase was 0.08 mm what means 1% diameter growth of branch. In 2019 the largest value of growth was 1.11% (0.05 mm) measured on June 25 between 6 pm and 7 pm. Our research was realized from May 28 to June 26 in years 2018 and 2019.

Considering to the values above we observed changes in branch diameters, as s reaction to the natural conditions. From the perspective of the irrigation is necessary complete data of monitoring whole vegetation period of plants samples without additional irrigation and plants samples with additional irrigation.

Key words: *dendrometer, precipitation, irrigation dose*

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Is there natural regulation of allergic common ragweed (*Ambrosia artemisiifolia* L.) pollen in the field conditions?

Patrícia MÁČAJOVÁ, Peter TÓTH

Supervisor: Assoc. Prof. Peter Tóth, PhD.

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. 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. Ragweed's dominate in disturbed habitats and tends to spread rapidly and negatively affect populations of indigenous plant species within ecosystems. The main goal of this study was to find ragweed plants, which making no pollen at all. The plants show very typical symptoms, young tissues are pale green/yellow, proliferation, malformation and virescence of male inflorescence, witches' broom, phyllody and stunting of the plant but plants stay still green/alive. The deformed male inflorescence forming "seeds" instead of pollen. Preliminary explorations were carried out across different habitats and ecosystems in Southwestern Slovakia from beginning of August until end of September 2019. The symptomatic plants were recorded and collected; we found their presence at 7 sites out of 12 sites visited. We assume that eriophyoid mites (Acari: Eriophyoidea) together with their symbiont bacterium-like organism called a phytoplasma (*Candidatus* genus Phytoplasma) are responsible for these symptoms. Until now we determined one eriophyoid mite from genera *Aceria*. It is the first report of this eriophyoid mite associated with common ragweed from Slovakia. The detection and exact determination of phytoplasmas will be performed by molecular techniques. Based on our survey we also estimate that the occurrence of these two organisms depends on habitat, with the preference of warmer areas and sandy soils. More robust data from field explorations supplemented by laboratory experiments will be required to obtain more powerful outputs. It is a high expectation that eryophyoid mites has a potential to become biocontrol agents.

Key words: *eryophyoid mites, excessive branching, phyllody, phytoplasma, male flower malformation, witches' broom*

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Determination of longitudinal dispersion at the Okna river

Martin MANINA, Peter HALAJ

Supervisor: prof. Ing. Peter Halaj, CSc.

Water pollution accidents occur frequently and cause severe damage in recent years. Freshwater is a scarce and valuable resource – one that can easily be contaminated. Once contaminated to the extent it can be considered "polluted," freshwater quality is difficult and expensive to restore. Longitudinal dispersion is a complicated transport process, which is very responsive to any hydrological, hydraulic, and vegetational change. The aim of this study is to represent an opportunity for determining a value of longitudinal dispersion at the Okna river. A river has a total length of 37 km, localized on the East Slovak lowland and it is a left – side tributary of Cierna Voda. Cross section changed by the natural morphological processes had original trapezoidal shape with bed width $b = 2\ m$, $h = 2.5$ and slope of the banks is 1:1. The measured flow ranged between 0.224 $m^3.s^{-1}$ and 0.311 $m^3.s^{-1}$. Mean annual discharge is $Q_a = 854\ l.s^{-1}$. Roughness coefficient has been determined on the base of field measurement $n = 0.045$. Longitudinal bed slope of our selected part of the river with total length of 100 m was $S_o = 0.012$. Average water depth during our measurements were from 0.3 m to 0.75 m and water temperature was 15.2 °C. We divided our interested part of the river to 3 cross sections of distances 0 m, 60 m and 100 m. Like a pollutant we used a NaCl tracer, which represented increasing values of conductivity in the water. On the base of the measured data's we determined a coefficient of longitudinal dispersion by the HEC – RAS and we calculated a dispersion by the equation:

$$C(x,t) = \frac{M}{2A\sqrt{\pi D_x t}} \exp\left(-\frac{(x - v_x t)^2}{4 D_x t}\right) \tag{1}$$

where: $C(x,t)$ is a mass concentration ($kg.m^{-3}$) in a place and time, Dx is the longitudinal dispersion coefficient ($m^2.s^{-1}$), A is a discharge area in stream cross section (m^2), M is a mass of a tracer (kg), x is a distance (m), t is a time (s), v_x is a mean flow velocity ($m.s^{-1}$) and compare these two theories. By the concentration curve we converted a measured conductivity to concentration of NaCl. Predicted longitudinal dispersion by the HEC – RAS were $0.237 – 0.311$ $m^2.s^{-1}$ and by the equation $0.027 – 0.033\ m^2.s^{-1}$. From the outputs of both theories we can say that model HEC – RAS approximated a measured data's very good what can be affected by the opportunity of model to include a factor of vegetation at banks and in the water, presented by the manning roughness coefficient. So many authors represented and used an equation (1), which is one of the possible tools how we can determine longitudinal dispersion but comparing with simulation with model HEC – RAS we can say that model is more than suitable for determination and can very sensitive predict a dispersion, even including vegetation in the river. By the water analysis in our laboratory we obtained that, water is three more times over polluted with total organic carbon, what can by consequence of fundamental processes of organic matter.

Key words: *concentration, HEC – RAS, dispersion, field measurements, equation*

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Soil volumetric water content measurement by gravimetric and TDR methods at field experiment Dolná Malanta

Lucia TOKOVÁ, Dušan IGAZ, Elena AYDIN

Supervisor: prof. Ing. Dušan Igaz, PhD.

Measurement of soil water content is fundamental procedure to many investigations in agriculture, horticulture, ecology, forestry, hydrology, waste management and other environmental fields. Methods of soil water content measurement can be divided into direct – gravimetric methods and indirect methods that are based on the measurement of another soil property which is dependent on soil moisture. The gravimetric method is the standard reference method against which other techniques are usually calibrated. The main aim of the presented paper was to compare measurements of volumetric water content by gravimetric and time domain reflectometry (TDR) methods in the field experiment at Dolná Malanta (Slovakia) during growing season of maize in 2017. Another goal was to perform a calibration on a particular device HydroSense II that was used in this study. The field experiment was conducted on four variants: control variant without biochar and N fertilizer (B0 + N0), variant with biochar at dose 20 t.ha^{-1} without N fertilizer (B20 + N0), variant with biochar 20 t.ha^{-1} and N fertilizer 160 kg.ha^{-1} (B20 + N160) and variant with biochar 20 t.ha^{-1} and N fertilizer 240 kg.ha^{-1} (B20 + N240). We have found that in variants with added nitrogen fertilizer (B20 + N160 and B20 + N240) the HydroSense II measurement accuracy operating under the TDR principle was lower when compared to the gravimetric method. TDR is nowadays well established dielectric technique to measure volumetric water content however its accuracy is influenced by high concentration of salts in the soil. We assumed that the higher measurement inaccuracy in the variants with nitrogen fertilizer may be caused by increased salt concentration in the soil as a result of applied N fertilizer.

Key words: *gravimetric method, TDR method, nitrogen fertilizer*

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Methodological issues of landscape-transforming analysis in adaptive inland water management

Dalma VARGA

Supervisor: Nóra Hubayné Horváth, PhD.

Hungary is located in the deepest part of the Pannonian Basin, which is exposed to water damage – especially lowland areas that are endangered by floods or inland waters. Several natural and anthropogenic processes contributed to the formation of inland waters after the period of river control (decisively from the middle of XVII. century) and still influence it. According to data from researches in 2015, 1.8 million hectares plain is endangered by waterlogging flooding. (Bozan 2015.) The aim of my research is to reveal landscape-transforming processes within waterlogging areas or areas endangered by inland water in order to establish adaptive inland water management methods. The analysis covers the study of former dominant land uses and linear infrastructure elements (green, blue and gray infrastructure) that can be determined. As a result of this analysis, the landscape-forming role of watercourses and the process of human interference can be observed and supported with maps including data. We can analyze land use characteristics of the past 200 – 250 years using historical, military maps, aerial photos. During these analyses it can be revealed, what type of land use was dominant within waterlogging areas in those times. The processing consisted of GIS methods: historical maps were georeferenced, the well-separable land uses with specific marks were digitalized. The results confirm the tendency of green surfaces, cultivated areas, the spread of settlements, in brief, the process of landscape-transform. Natural watercourses and artificial trenches, canals, drainage systems can be determined as linear elements in the landscape, likewise road network and linear green infrastructure elements such as alleys, lanes with trees and rands. In our research, analyses are based on historical maps, aerial photos, comparing different periods of 50 – 60 years prevalence. It's easier to define surface-like elements on historical maps, they give completely accurate data of contemporary conditions. The same cannot be said for linear infrastructure elements. This is especially true in the case of watercourses, since in many cases several areas can be determined without clearly identified water network. We can get informs about these watercourses – such as canals, drainage systems – from additional registers, data. As a conclusion, landscape-transforming analyses provide valuable data for defining land use changes, which complemented with hydrological databases – especially in waterlogging areas – adequately supports adaptive inland water management methods, that are the most adapted to the specific characteristics of the area.

Key words: *inland water, landscape-transforming processes, green infrastructure*

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Contamination of soil by mobile lead fraction

Jozef VARGA

Supervisor: doc. Ing. Radoslava Kanianska, CSc.

Lead is one of the best known potentially toxic elements in the environment. Its presence in soil can contribute to the food contamination and serious health risks. Soil can accumulate Pb also from anthropogenic sources including mining industry. The aim of the study is an evaluation of the Štiavnica river alluvium contamination by the mobile lead fraction. We also evaluate the influence of some soil properties on the content of mobile lead fraction in soil. Štiavnica river flow through Štiavnica Mountains and Krupina Plain. Registered environmental burden Lintich (EBL) is situated at the upper part of the stream. We selected six study sites of which one is represented by EBL (2EBL). Others five study sites are under different land-uses and they are located at river alluvium (A). The first study site (1A) is situated above EBL and others (3A-6A) downstream bellow EBL. We applied the first step (solution A – acetic acid- HOAc 0.11 mol/l) of the sequential extraction procedure (modified European Community Bureau of Reference - BCR procedure) to determine the mobile lead fraction (water soluble, exchangeable and bound to carbonate) and thus the most available fraction able transfer from soil to plant. We measured in soil samples pH, Eh, soil organic carbon (SOC) and clay content. The content of Pb as HOAc extractable fraction varied among study sites from 0.24 to 185.70 $mg.kg^{-1}$. The highest content was measured at 2EBL study site, the lowest content was measured at 3A study site. All measured data exceed limit value for Pb (set up as a fraction possible to transfer from soil to plant) according to the Decree no. 59/2013. Limit value for Pd determined in 1M NH_4NO_3 solution is 0.1 $mg.kg^{-1}$. Among selected study sites are two (5A, 6A) used as arable land where is risk of agricultural production contamination by lead. These study sites are located at the Krupina Plain, the region intensively used for agricultural purposes. Its contamination mainly at river Štiavnica alluvium is probably caused by transition of heavy metals from Štiavnica Mountains resulting from past mining activities. We also observed the relations between higher mobile lead fraction, clay and organic carbon content in soil.

Key words*: soil contamination by lead, mobile lead fraction, environmental burden, soil property*

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SECTION

Plant Production

Synthesis and cytotoxic activity of galactomannan-gemcitabine conjugates

Dilnoza AMONOVA, Bahtiyor MUHITDINOV, Husniddin KIRGIZBAYEV

Supervisor: Academician, Prof. Dr. Abbaskhan Turaev

Galactomannans are naturally occurring heteropolysaccharides composed by a β-(1,4)-D-mannan backbone with a single D-galactose branch linked by α-(1,6). These biopolymers structurally differ from each other with respect to the mannose/galactose (M/G) ratio. Galactomannans are usually isolated from the endosperm of dicotyledonous seeds of numerous plants. The M/G ratio varies with plant origin but is typically in the range of 1.0 – 1.1 : 1.0, 1.6 – 1.8 : 1.0, 3.0 : 1.0, and 3.9 – 4.0 : 1.0 for fenugreek, guar, tara, and locust bean gums, respectively. Galactomannans are major polysaccharides of commercial importance in various industries due to their low cost and excellent viscosifying properties. Galactomannans are extensively used in food industries as stiffeners and stabilizers of emulsions for preparation of dietary products e.g. coffee whiteners, baby milk formulations, seasonings, sauces and soups, tinned meats and frozen and cured meat foods. In addition, these biopolymers are largely used in oil recovery, pharmaceutical formulations, and personal care products. Recent studies showed the galactomannans could be used to prepare films and hybrid material for biomedical applications. In addition, galactomannans are promising biomaterials for the drug delivery, due to their abundance in nature, low-cost processing, biocompatibility, biodegradability. These polysaccharides have a large number of reactive hydroxyl groups on their backbone, which can be easily modified chemically to many types of derivatives.

In the present report, results regarding the synthesis and cytotoxic activity of galactomannan-gemcitabine conjugates were abstracted. In the studies, low molecular weight galactomannan with weight average molecular weight (Mw) of 40 kDa (G/M = 1.0/2.3) was prepared by acid assisted depolymerization of commercial guar galactomannan (Mw = 563 kDa, G/M = 1.0/1.8). The galactomannan prepared was periodate-oxidized with sodium periodate (1.0 : 0.3 mol : mol) for 24 h. The obtained dialdehyde galactomannan was then dissolved in water and reacted with gemcitabine hydrochloride for 24 h following precipitation and washing with ethanol. The final reaction product was purified by dialyzing and freeze-dried. Chemical structure of the galactomannan-gemcitabine conjugate synthesized was approved by IR- and NMR spectroscopic methods. The studies showed the gemcitabine had mainly conjugated by C2/C3 carbon atoms of mannose residues in galactomannan backbone by imine bond formation. The drug content (1.64%) and Mw (32.5 kDa) of the conjugate were determined by UV spectroscopic and gel permeation chromatography methods, respectively. The cytotoxic activity assays were carried out with mouse C127 (breast cancer) cells. In the results, the galactomannan-gemcitabine conjugate exhibited cytotoxic activity of 48.0 – 87.9% with dose-dependent manner compared to the free drug. The results indicate the galactomannan-drug conjugate presents high cytotoxic effect despite the much less content of the freed drug. Thus, the galactomannan-gemcitabine conjugate has perspectives as an antitumor prodrug candidate and further trials required to develop the antitumor prodrug with less adverse effects.

Keywords: *galactomannan-gemcitabine conjugate, synthesis, cytotoxic activity*

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Phenotyping of wheat varieties with different drought tolerance

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Supervisor: prof. Ing. Marián Brestič, CSc.

In recent years, climate extremes (drought and high temperature) have been a significant threat to the production properties of crops in the Central European region, not only in the southern regions of Slovakia, but also in higher altitudes. Therefore, in terms of plant physiology, it is extremely important to address the responses and properties of plants after extreme events. In this respect, it is essential to have appropriate physiological criteria to enable more efficient and faster selection of genetic resources and their tolerance properties to environmental stress. The purpose is to characterize changes that are related to production properties and to address them as new approaches potentially usable in breeding. The aim of the work was to characterize specific phenotypic manifestations of genetic resources and modern varieties of wheat, their tolerance in various environmental conditions (under the influence of drought and high temperature), subsequently to characterize changes in behavior of studied genotypes and varietal differences in growth-production properties, at the level of various physiological and biochemical parameters. Experiments were carried out in a greenhouse under transparent foil with 35 genotypes of summer wheat winter form (*Triticum aestivum* L.) with various ploidyties and dry tolerance. Modern varieties with different anthocyanins in the grain were also used to determine tolerance. The plants were grown in plastic pots and divided into two variants. First variant was controlled with sufficient watering and second variant was stressed by drought with the application of irrigation regime (reducing the water dose to 1/3 of the total dose of the control variant). During the summer season, control and stressed variant plants were monitored and measured 2 times per week. Non-invasive methods were used to identify the physiological properties of plants, for the evaluation of chlorophyll fluorescence parameters characterizing the structure and state of the photosystem 2 (PSII) (Handy Pea, Hansatech, GB), specific photosynthetic parameters derived from measurements of absorbance and fluorescence (PhotosynQ, USA), as well as water-mode parameters, such as the relative water content (RWC) of the leaves. All measurements were focused on the ability to acclimate wheat plants. Using tens of measured parameters, our analyses point out that the drought-stressed variant had relatively low PSII reaction center values as well as low electron transfer rate (ETR) values compared to the control variant. Specific plant responses indicate that control variant had a better relative state of osmotic dry leaf adaptation (90 – 96%) compared to the stressed variant (60 – 85%). The results also show the possibility of using selection criteria potentially useful in more extensive screening of genetic resources.

Key words: *wheat, tolerance, drought, photosynthesis, chlorophyll fluorescence*

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Utilisation of repellents for the control of selected plant pests and evaluation of their effect to non- target insects

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In our experiment, there were tested insect repellents, which are based on chemical products with an offensive smell or taste and essential oils (EOs). The most effective and the most known insect repellents currently on the market are DEET (N, N-diethyl-3-methylbenzamide) and 2-Undecanone. At present, botanical pesticides are at the forefront in plant protection and agricultural pest management and they are the best suited for the use in organic food production in industrialized countries. EOs of aromatic plants are effective natural products as contact and fumigant insecticides and as repellents against stored food pests.

The aim of the study was to find the level of repellent effect of the chemicals and essential oils on the Western corn rootworm (*Diabrotica virgifera virgifera*). The flight of the adults started at the beginning of July. In the experiment, there were used four different products with the concentration of 1%. Chemical products DEET, 2- Undecanone, EOs of garlic (*Allium sativum*) and eucalyptus oil.

One liter of the repellent spray was used at the area of 2 x 3 meters. After 24 hours and 48 hours it was checked the repellent effect. In the case of both applications, it was 30 °C and breeze. After 48 hours the numbers were different and the temperature was a little bit colder, and the insect was hidden, at the bottom, of leaves.

Average number of beetles on one plant after 24 hours depended on the variant and it was as follows: Undecanone 0.36; DEET 0.64; Garlic 0.88; Eucalyptus 1.12 and the control 1.12. After 48 hours the average number of beetles was in Undecanone 1.12; Eucalyptus 1.2; DEET 1.24; Garlic 1.32 and 1.88 in the control variant.

It was found significant effect of Undecanone application compared to control variant (ANOVA, LSD multiple range test at 95.0% level).

Key words: *DEET, 2 - Undecanone, Alium sativum, Eucalyptus oil, Diabrotica virgifera virgifera)*

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The analysis of antioxidant activity of ramson (*Allium ursinum* L.) – inflorescens and bulbs

Michaela HRÚZOVÁ

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The ramson (*Allium ursinum* L.), named also as wild garlic belongs to perspective species for agri-food and for phytotherapeutical uses. Antioxidant activity of ramson dry inflorescences and bulbs were analysed. Samples were prepared from dry inflorescences (locations Štrkovec, Sikenica and Šoporňa) and from dry bulbs (location Sikenica). Antioxidant activity was determined by DPPH (2.2-diphenyl-1-picrylhydrazyl-hydrate). Extracts were prepared from 1 g of substance and 25 ml of methanol (99.50%) and distilled water, mixing on the shaker during 12 hours. For basic solution the 0.025 g of DPPH and 100 ml of methanol were used. The measurements were carried out by a spectrophotometer Genesys 20, model 4001/1 at the wave length 515 nm. The decrease is recorded after 10 minutes of reaction. The antioxidant activity of ramson inflorescences in methanolic extracts was in the range from 12.85% to 20.63% and in aqueous extracts from 14.78% to 24.97%. In ramson dry bulbs methanolic extract was the antioxidant activity 14.95% on average and in water extract 19.72% on average. The strongest antioxidant power had the samples of inflorescences from the location Šoporňa in water extract (22.20%) and the location Štrkovec in methanol extract (19.74%). The inflorescences samples from location Sikenica had lowest activity on average in water extract (17.07%) and methanol extract (13.70%) too. In both cases (dry inflorescences and dry bulbs of ramson), the antioxidant activity is low.

Key words: *Allium ursinum, antioxidant activity, extrats, inflorescences, bulbs*

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Exogenous effect of abscisic acid in the system of antioxidant protection and biochemical characteristics of cotton-plant under saline stress

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During the research on the effect of phytohormone of abscisic acid under abiotic condition of vegetation some regularity for the formation of cotton plant resistance to saline effect was defined in resistant (Gulistan) and sensitive (C-4727) varieties of cotton plant seedlings of *Gossypium hirsutum* L. that is differentiated by saline resistance. Experiment was realized in laboratory condition. The material of the study is 7-day-old seedlings of tolerant cotton varieties Gulistan and stress-sensitive varieties C-4727. The seedlings were grown in tap water, then the samples were subjected to salt stress by exposing them in a solution of 1% and 4% sodium chloride and ABA (consentration of ABA 10^{-7}M) for 1 hour, and then for 24 hours, followed by determination in the seedlings of the activity of APO, SOD, contents of proline and MDA. Also conducted comparative research on antioxidant potential of cotton plant varieties that are different in saline resistance: superoxide dismutase, ascorbate peroxidase, also malonic dialdehydes and free amino acid proline. The content of malonic dialdehydes is lower in saline resistant variety, while the activity of ascorbate peroxidase enzyme is higher compared to stress sensitive varieties of cotton plant. The research showed a significant difference of sensitive and resistant cotton plant varieties in the reaction to saline stress. The difference in the reaction to salification connected with the difference in enzyme activity of antioxidant system, free amino acid proline content and malonic dialdehydes. Phytohormone ABA influences on antioxidant system of protection in the effect of saline stress that consequently increases cotton plant resistance to salification. The conducted research showed high constitutive level of free amino acid proline and ascorbate peroxidase for the resistance of the variety Gulistan.

The data on the influence of phytohormone ABA on the functioning of antioxidant system of protection of cotton plant in a normal condition and in the effect of oxidative stress holds a considerable importance for the knowledge of mechanisms of coordinated regulation of components of growing part response in the process of realization of adaptive program.

This fundamental knowledge can be used in elaboration of technology for creating intragenic plants with high resistance to abiotic stresses.

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The role of phytohormones in the regulation of cotton resistance to the action of chloride salinity

Karomat M. KULDOSHOVA

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Soil salinity is a major abiotic stress in agricultural crop productivity worldwide. Salt stress has a significant effect on plant growth and development. Under salt treatment, the seed germination, root length, plant height, and fructification of plant are significantly inhibited. Osmotic stress is the first stress experienced when a plant is exposed to saline soil, and it instantly affects plant growth. Ion toxicity occurs later when salt levels reach a threshold, beyond which the plant cannot maintain ion homeostasis and growth. Ion toxicity and osmotic stress are primary stresses that can cause oxidative stress and a series of secondary stresses. Salt stress also leads to a decrease in photosynthesis and results in a substantial decrease of crop yield worldwide. Adaptation of plants under stress conditions is manifested in changes in physiological and biochemical processes, and these changes are controlled by the cell genome by repression and derepression of individual genes. The formation of cotton resistance to unfavorable abiotic factors is a complex, multicomponent process, including both specific and general (non-specific) reactions. The latter include, in particular, changes in the balance of phytohormones. Some of them, and, above all, the stress hormone - abscisic acid (ABA) is able to act as a regulator, determining the response of plant cells to adverse effects. Not enough attention is paid to the participation of phytohormones in increasing plant resistance in the initial period when the unfavorable factors affect it, although the results of a number of studies indicate that it is during this period that very important, if not the main, events occurring in cells and tissues of a plant organism, largely predetermining the entire the subsequent course of the formation of resistance. Indole-3-acetic acid (IAA) – a plant hormone plays a major role in regulating plant growth and development. Its accumulation, defined as auxin maxima, is well correlated with newly generated organs of plants. For example, it essentially contributes to vascular tissue development, cell elongation, organogenesis and apical dominance by auxin maximal formation. The work was initiated to investigate the response of cotton with the participation of phytohormones (ABA and IAA) to the effect of abiotic factors, and to examine some mechanisms for increasing cotton resistance to salt stress. In consequence of the above, we studied the participation of the phytohormones ABA and IAA in the adaptation of cotton seedlings to abiotic factors. Based on the qualitative and quantitative analysis carried out by the HPLC method, it was found that the content of IAA and ABA in seedlings is different, indicating a different function of each phytohormone. The determination of endogenous phytohormones under the influence of abiotic factors revealed their different content in the dynamics of the development of seedlings resistance of the cotton variety Gulistan to salinization. It is established that in the initial period of stress after 1 hour, the content of ABA increases dramatically (on average by 200%), while IAA decreases slightly (10 – 15%).
Studying the effect of phytohormones on enzyme activity under stressful situations is of great scientific and practical interest. Therefore, we further investigated the enzymes of antioxidant systems in seedlings under the action of ABA and IAA.

Keywords: *abscisic acid, indole-3-acetic acid, salinity, phytohormone*

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Effect of harrowing on nitrogen content in yield of mixture of spring triticale and faba bean

Agnieszka ŁAGOCKA

Supervisor: dr hab. inż. Piotr Sobkowicz, prof. nadzw.

Mixtures of legumes and cereals become increasingly important in recent years. Both as feed with a really high protein content for farm animals and as a crops that are leaving good positions for further crops, especially cereals. By using leguminous-cereal mixtures, their biodiversity makes it possible to eliminate the harmful effects of successive cereal crops. On the other hand, the phytosanitary action of the leguminous component allows to free the field from many soil pathogens. Unfortunately, the problem that can occur in the cultivation of mixtures is the problem of weed infestation. Currently, there are no registered weed control substances on the market that are permitted for use in both field cultivation, e.g. field beans and spring triticale. At the same time, existing sustainable management systems require that the use of chemicals should not be the only method of regulating weed infestation in crops. The intention is also to reduce the penetration of chemicals into the environment. For these reasons, the basic method of destroying weeds in mixtures is to harrow the crops with a weeder harrow. This procedure, carried out at the proper stage of the mixture growth, successfully destroys weed seedlings, especially spring weeds, such as white compost, which was the most common weed in the studied mixture. The experiment investigated the effect of harrowing on the percentage nitrogen content in triticale grain and field bean seeds. The experiment was a field experiment and was conducted for a period of three years at the Swojec experimental institute belonging to the Wrocław University of Environmental and Life Sciences. The plants for testing were harvested from each experimental plot at full maturity. In properly prepared samples, total nitrogen was determined using the Kjeldahl method. The observations showed that the treatments did not significantly affect the nitrogen content in the mixture components. In each of the objects the N content was maintained at a constant level. In the case of triticale seeds, most nitrogen was shown in the mixture, which was harrowed once deep in the second term of harrowing. On the other hand, in faba bean seeds, the highest amount of nitrogen was observed after increasing the sowing rate of the mixture. The seeds of faba bean were characterized by the lowest amount of this element, which in the leguminous-cereal mixture was not treated with any protective treatments against weeds. The mixture, which was harrowed once, shallowly in the third term was characterized by the lowest amount of nitrogen in the grains of the grain studied. However, none of these results were statistically confirmed.

Key words: *harrowing, nitrogen, faba bean, spring triticale, mixtures*

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Influence of different intensity of mowing on grassland botanical composition in warm and dry conditions

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Supervisor: doc. Ing. Ľuboš Vozár, PhD.

The aim of the work was to find out the effect of different mowing intensity on the grassland. The experiment was implemented in the village of Žirany, which is characterized by a dry and rainfall with annual average temperatures 9 °C. The soil type of fluvisol prevails with a weakly acidic to acidic soil reaction at the locality. Permanent grassland was used for sheep grazing prior to the experiment. From the floristic point of view, grasses predominated and the largest species present is *Lolium perenne* L. Monitored grassland was divided into 3 variants and fertilized with the same nutrient dose N (60 kg.ha^{-1}), P (40 kg.ha^{-1}), K (80 kg.ha^{-1}). Grass vegetation was cut in individual variants as follows: Variant 1 – mowed 3 times (1st mowing at the time of hay-mowing maturity, 2nd mowing after 60 days from first mowing and 3rd mowing after 60 days of mowing second), Variant 2 – mowed 2 times (1st mowing in hay-mowing maturity, 2nd mowing after 90 days since first mowing) and Variant 3 – mowed 1 times (1st mowing at the time of seed maturation). We evaluated the floristic composition of the stand before each mowing. Floristic evaluation of each stand was done by the Reduced Projective Dominance method. According to the Shannon Index we have calculated the diversity of grassland. The monitoring period was 2017 and 2018.

The highest proportion of all observed botanical species had grasses. In all the monitored variants, the proportion of grasses per year increased, which was influenced by the different intensity of exploitation. We evaluated the highest difference on variant 1, which was cut 3 times, where the average proportion of grasses increased from 29.7% (2017) to 84.7% (2018). Especially *Lolium perenne* L. dominated among the grasses, which in 2017 had the highest part among grasses in variant 2 in 2nd mowing (32.5%). The proportion of leguminous decreased in 2018 compared to 2017. On year's average, we found the highest difference in variant 1, where the average share of leguminous substances decreased from 30.2% (2017) only to trace amount (2018). The lowest decrease was in variant 2, where the average proportion of leguminous substances decreased from 10.8% (2017) to 0.5% (2018). *Trifolium repens* L. in variant 2 in the 2nd mowing (31.0%) had the highest share of individual species of leguminous in 2017 and trace amount in 2018.

Other meadow herbs compared to leguminous plants had a higher proportion in the monitored variants. The highest difference in the annual average of weights was found in variant 1, where the average value of herbs decreased from 28.5% (2017) to 5.8% (2018). The highest proportion of other grasses had *Achillea milefolium* L. in 2017 in variant 1 and 3rd mowing (38.0%) and in 2018 its share decreased to 12.5%.

At the beginning of the follow-up (2017), the diversity index ranged from 1.42 to 2.19. The lowest value (1.42) was found on variant 2 after the 2nd mowing (2017). Variant 3 (2.19), where only one mowing was done during the year, achieved the highest average diversity value. In individual variants in next year (2018) after one cut compared to 2017 values decreased but variants that have been cut more than once had a higher average diversity value.

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Impact on yield forming parameters, seed yield and oil content in seeds of sunflower by using plant growth stimulators

Ján MAREK

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The aim of the field poly-factorial experiment was to evaluate the effect of two plant growth stimulators, Florone and Fertisilin, on selected yield-forming parameters (number of plants, number of heads, head diameter, weight of head, weight of thousand achenes), seed yield and oil content in seeds of sunflower. All mentioned parameters were observed and analysed on sunflower hybrid 'SY Gracia', which has a high yield potential and can be cultivated with ClearField plus technology. The experiment was established at the research field of the Plant Biology and Ecology Centre of the Faculty of Agrobiology and Food Resources of the Slovak University of Agriculture in Nitra – Dolná Malanta in vegetation season of year 2018. The plant growth stimulator Florone is a product on free amino-acids basis with added NPK and microelements such as B and Mo. The stimulator Fertisilin contains, in addition to microelements, a silicic in the form of ortho-silicic acid, which is fully accessible and assimilated by plants. Both stimulators were applied, depended on variant, in three terms (phase of 6-8 leaves, beginning of flowering, and both terms). Dosing of stimulator Florone was 0.4 l.ha^{-1} and Fertisilin 0.7 l.ha^{-1}. The influence of biostimulators on the number of plants (59 578 pcs.), number of heads (59 667 pcs.) and yield of sunflower (4.24 t.ha^{-1}) was statistically non-significant. The influence of biostimulators on head diameter (244.1 mm), weight of head (194.46 g) and weight of thousend achenes (73.45 g) of sunflower was statistically significant. The influence of plant growth stimulators on oil content in achenes (43.85%) was evaluated as statistically high-significant. By using of statistical variance analysis ($\alpha = 0.05$), the influence of application of plant growth stimulators on number of plants, number of heads and seed yield of sunflower was not determined. Conversely, the influence of plant growth stimulators on head diameter, weight of head, weight of thousand achenes and oil content by using of variance analysis ($\alpha = 0.05$) was determined. The highest values of head diameter (261.6 mm), weight of head (269.75 g) and weight of thousand achenes (86.5 g) were observed in variants treated by plant growth stimulator Florone. The highest values of seed yield (4.48 t. ha^{-1}) and oil content (45.81%) were observed on variants treated by plant growth stimulator Fertisilin.

Key words: *sunflower, biostimulators, yield-forming parameters, yield, oil*

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Impact of different caespestechnical level for development of the turf root system

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The root system of grasses is one of the most important parts of the plant that affect the quality and vitality of grass biocoenoses. A significant part of roots (88 – 95%) is situated at a depth up to 100 mm. The maximum depth of penetration of grass roots into the soil is in case of the majority of grasses 1 – 1.5 m. In case of some dry resistant grasses it is 2.5 – 3 m. The life cycle of roots is very short, max. 1.5 years. New offshoots formed after loosing older parts of grass form their own root system (Holúbek et al., 2007; Vozár, Jančovič, 2011). Jančovič (1997) writes that the production of underground biomass is connected with some limiting factors. The underground parts represent about 50 – 80% of the total vegetable matter during the period of maximum growth and more than 90% of the vegetable matter in spring and autumn. With increasing production of crop because of nutrition the relative accumulation of assimilates into the roots is decreasing. The roots that are older contain suberin, the roots stop to absorb water and become storage roots. Suberin is a corky substance. These variations can be corrected by soil conditioners (Jančovič, 1997). The aim of the experiment was therefore to examine the possibility of using selected soil conditioners for development of the turf root system. The Experiment was carried out in the area of Oponice, district Topoľčany between the years 2016 and 2018. The area climate is characterised as warm, slightly humid, with mild winters. The average daily temperature per year is 7.4 °C, and during the vegetation period 13.6 °C. The average annual rainfall is 593.0 mm, during the vegetation period 338.0 mm. In this article we are presenting the results after three years of land management. To examine the impact of soil conditioners with NPK fertilisation, a small-lot experiment in raised wooden boxes with dimensions 2 x 5 m was established. The boxes were filled with quartz sand marked as SH 34 of pH 7 with fraction 0.1 mm – 0.4 mm. A grass mix of the species *Lolium perenne* L. and *Poa pratensis* L. was used. The seeding amount was 25 $g.m^{-2}$. We were observing 6 variants in total: Variant A: Alga 600 was applied 2 times per month by spraying on the leaves in a dosage of 0.5 $kg.ha^{-1}$, Variant B: RootMost applied in a dosage of 1 $l.ha^{-1}$ in a 10 days interval by spraying on the leaves, Variant C: Vulkagran G ® was during establishing of the experiment applied once in strips in dosage of 100 $g.m^{-2}$, Variant D: Hack Bazalt 100 g/m^2, applied once in strips during establishing, Variant E: Agrosil® LR was applied once during establishing of the experiment in a dosage of 100 $g.m^{-2}$, Variant F: control sample – unfertilised variant. After three years of research (2018) it was found, that in sandy conditions, the largest root system was in case of application of the combination Agrosil LR + NPK (457.36 $g.m^{-2}$). Only slightly lower root formation was present in a variant with the fertilisers Root most + NPK (418.10 $g.m^{-2}$). The lowest production of roots was paradoxically found in case of combination of fertilisers Vulkagran G ® + NPK with the value of (288.47 $g.m^{-2}$) and almost the lowest production of roots was in case of control sample (289.65 $g.m^{-2}$). The stubble also represents an important reserve part (Holúbek et al., 2007). The highest value was found in case of combination of Agrosil® LR + NPK (1189.49 $g.m^{-2}$). The impact of conditioners was manifested fully, and the provably lowest weight had the stubble in the control sample without the application of conditioners (655.58 $g.m^{-2}$). The pH values of soil were between 6.77 in variant C and 7.16 in variant B which is classified as neutral pH. The cation exchange of soil reached values between 80.85 $mmol(p^+).kg^{-1}$ in variant B and 108.30 $mmol(p^+).kg^{-1}$ in variant C.

Key words: *turf, soil conditioners, fertilisation, roots, variants*

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The efficacy of fungicides against Fusarium head (and grain) blight under natural infection

Ľudovít MIŠĽAN

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Fusarium head blight (FHB) is a dangerous cereal disease that causes direct crop loss and mycotoxin contamination of grain. Protection against disease is a complex of measures, including fungicidal protection. Due to the complicated pathosystem, reliable chemical protection is still not solved in practice. The aim of this work was to evaluate the efficacy of authorized fungicides on the occurrence and harmfulness of this disease under conditions of natural infection. Small plot field trial was realized on wheat, variety Midas, in the locality Pavlovce nad Uhom (land owner - "Gama" company). The fungicides Capalo, Amistar Opti, Prosaro 250 EC and their combinations were used, which were applied in the T1 – T3 term (BBCH 32, 59, and 61). Evaluation of the affected ears was performed on 100 randomly selected ears to be repeated, determining the % of infected area of the ear by FHB. The fungicidal efficacy (FU) was calculated according to the Abbott formula. The average damage by FHB in the control variant was 27.91%. The fungicidal efficacy was determined as follows: Capalo (T1) – 15.62%, Amistar Opti (T2) – 18.92%, Prosaro 250 EC (T3) – 46.75%, Capalo + Amistar Opti (T1 + T2) – 31.55 %), Amistar Opti + Prosaro 250 EC (T2 + T3) – 59.44%, Capalo + Prosaro 250 EC (T1 + T3) – 57.74%, Capalo + Amistar Opti + Prosaro 250 EC (T1 + T2 + T 3) – 72.44 %. The average grain infestation in the untreated control was 9.5%. The fungicidal efficacy evaluated on the basis of the grain infestation was as follows: Capalo (15.79%), Amistar Opti (31.58%), Prosaro 250 EC (53.16%), Capalo + Amistar Opti (55.26%), Amistar Opti + Prosaro 250 EC (52.63%), Capalo + Prosaro 250 EC (73.68%), Capalo + Amistar Opti + Prosaro 250 EC (57.89%). From the above results it is possible to sort the treatments in descending order as follows: on ears Capalo + Amistar Opti + Prosaro 250 EC, Amistar Opti + Prosaro 250 EC, Capalo + Prosaro 250 EC, Prosaro 250 EC, Capalo + Amistar Opti , Amistar Opti, Capalo; on grains: Capalo + Prosaro 250 EC, Capalo + Amistar Opti + Prosaro 250 EC, Prosaro 250 EC, Amistar Opti + Prosaro 250 EC, Amistar Opti, Capalo. The fungicide efficacy against FHB does not correspond to the efficacy against grain fusariosis. The results of the work show that the use of fungicides in the appropriate term of application (T3, BBCH 61) significantly affects the percentage of *Fusarium* infestation of ears and grains. However, no tested combination of fungicides provides complete protection against ear and grain fusariosis.

Key words: *winter wheat, ears, grains, Fusarium, fungicides, efficacy*

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The effect of the natural preparation DAG-1 on the catalase activity of cotton

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Environmental stresses cause oxidative stress as a result of water deficit. The plant cells display non-enzymatic and enzymatic antioxidant system to mitigate the oxidative damages caused by reactive oxygen species (ROS). In general, antioxidant activity of enzymes and scavenging of reactive oxygen species is increase due to salt stress to improve tolerance of a plant.Researchers at the Institute of Bioorganic Chemistry Uzbekistan Academy of Sciences has extensive experience in creating new generation drugs based on natural compound DAG-1 (Dalimov, Akhunov, Gafurov, as per title letters of the main authors) and studying the molecular mechanisms to biotic and abiotic stresses. The uniqueness of natural inductors consists in the ability to be active in low doses, and at the same time they are non-toxic both to humans and the surrounding nature. It should be particularly emphasized that they enhance the immune system of plants. The aim of this review is to determine the optimal concentration of DAG-1 from different doses (10^{-6}, 10^{-7}, 10^{-8} M) for biotechnological cotton species of Porloq-1 and Porloq-4 under laboratory conditions. As we know, each variety has individual biochemical parameters, and we must determine the optimal concentration for each variety in order to increase the stability of cotton. Based on this, we studied the catalase (CAT) activity increasing the tolerance of plant maximally. The CAT catalyses the decomposition of hydrogen peroxidase to water and oxygen. Each second one CAT molecule can convert millions of hydrogen peroxide molecules into water and oxygen. The CAT activity was determined by the Sinha method, in this method dichromate in acetic acid is reduced to chromic acetate when heated in the presence of undecomposed hydrogen peroxidase (H_2O_2), with the formation of perchromic acid as an unstable intermediate. The chromic acetate is measured calorimetrically at 570 nm. DAG-1 caused increase in the activity of CAT, and maximum increase for different doses (10^{-6}, 10^{-7}, 10^{-8} M) for the cotton variety Porloq-1 was found at 10^{-7} M and, the CAT activity was the 98.21 (unit mg-1 protein min-1), in the control (without DAG-1) the CAT activity 80.78 (unit mg-1 protein min-1). Maximum increment (30.00% of control) in CAT activity was observed at the 10^{-8} M dose for cotton variety Porloq-4. As a result, the CAT activity was 87.00 (unit mg-1 protein min-1) and in control variant the CAT activity showed 66.92 (unit mg-1 protein min-1). The obtained results showed, that, each variety requires an individual approach of using the DAG-1 preparation to increase of plant tolerance to adverse environmental factors.

Key words: *catalase, DAG-1, hydrogen peroxide*, *reactive oxygen species, cotton*

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Sowing time effect on formation of faba bean yield in 2018

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Faba bean (*Vicia faba*) is one of the most grown legumes in the world. The crop is mostly grown for seeds, who have high crude protein content, and are used for food and feed. In Baltic countries, only few resarch results about sowing time effect on faba beans' yield formation are documented. The aim of this study is to compare faba bean yield and yield components depending on three sowing times. The study was carried out at the Research and Study Farm "Pēterlauki" of the Latvia University of Life Sciences and Technologies (LLU) in 2018. Four factors were researched: A – sowing time (21, 29 April, and 08 May), B – variety ('Laura', 'Boxer', 'Isabell'), C – sowing rate (30, 40 and 50 germinable seeds m^{-2}), D – treatment with fungicide (with and without application of fungicide Signum (boscalid, 267.0 g kg^{-1}, pyraclostrobin, 67.0 g kg^{-1}), 1 kg ha^{-1}). Beans' yield (t ha^{-1}) was detected in the trial and yield components were determined using sample sheets (10 plants from each plot). Conditions were not suitable for high yield formation due to drought and high temperatures in 2018. The yield between first two sowing times (21 April and 29 April) did not differ significantly, but on average the highest yield (3.33 t ha^{-1}) and number of seeds per plant (24.12) was provided by sowing beans on 29 April. Average number of pods per plant (7.71) and seeds per pod (3.20) was higher when sowing beans on 21 April. Due to unsuitable meteorological conditions for bean developing and yield formation, beans sown in 08 May had the lowest yield (0.94 t ha^{-1}), number of pods and seeds per plant (7.60 and 23.44), and seeds per pod (3.09). This can be explained with irregular development of crop: we could observe ripened pods, green pods and even flowers on different branches of one plant. On average, the highest yield was provided using the variety 'Boxer' (2.64 t ha^{-1}); the best sowing rate was 50 germinable seeds m^{-2} (2.64 t ha^{-1}); and fungicide application gave yield increase (2.69 t ha^{-1}). Number of pods per plant (8.27) was higher for the variety 'Laura', but number of seeds per plant (24.93) and seeds per pod (3.34) was higher for the variety 'Isabell'. The best sowing rate for the number of pods per plant (8.29) and seeds per plant (25.93) was 30 germinable seeds m^{-2}, but for the number of seeds per pod (3.17) – 40 germinable seeds m^{-2}. Fungicide application did not give increase for any yield component.

Key words: *field bean, yield, seeds, pods*

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Change of soil properties of grassland by application of digestate

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The article presents a change of soil properties of grassland on the soil type of cambisol due to nitrogen fertilizer of organic origin - digestate. The substrate was created as a secondary production of biogas stations. In ecological management, it was used to maintain the fertility characteristics of soils of the terrestrial ecosystem, in order to ensure the nutrition of above-ground and underground biomass and the sustainability of soil qualitative properties.

The aim of the research task was to evaluate the impact of digestate application from the aspect of some non-production functions of the ecosystem. The evaluation of the change in soil properties was analysed in three years of utilization, used by three times mowed. The created sustainable management conditions were tested in relation to nitrogen nutrients in four variants. The output was the transfer of knowledge for practice. The digestate was applied by spring fertilization once to the grass surface. The research area is located in the mountainous part of Slovakia. Slight slope at an altitude of 480 m has a north-east exposure. The experiment with individual fertilization rates was established by the block method in three replicates on experimental plots (area of 10 m^2). Fertilization was applied on the basis of pure nitrogen nutrients (N) in $kg.ha^{-1}$, while variant V1 was not fertilized, variant V2 contained 90 $kg.ha^{-1}$ of N, variant V3 had an increased dose to 120 $kg.ha^{-1}$ of N, the highest dose of 150 $kg.ha^{-1}$ of N had V4. The change in soil properties after fertilization by the availability of soil nutrients were analysed by the indirect method of respirometric activity of microorganisms in the soil profile. The content of CO_2 production was determined by the Infrared gas analyser. The activity of microorganisms supported other biochemical processes and their activity confirmed the positive effect of the nutrient release process in plant-accessible (inorganic) form, which was also observed in the production of phytomass during the growing year. Changes in soil ability in terms of enzymatic activity of microorganisms, depending on the quality level of biological processes, were evaluated by "*in situ*" measurements on the stand surface (average values for 2016 – 2018 ranging from 489 to 505 ppm) and in the root zone in depths up to 100 mm profile (ranging from 489 to 547 ppm). Evaluation of the data revealed that in the first crop year higher enzymatic activity was recorded on the soil surface, in the second and third years the increased biological activity measured at a depth of 100 mm of soil profile. Within the variants, this activity was found in all fertilized variants. The correlation relationship was confirmed between fertilization and respirometric activity (r = + 0.6294 [++]), with statistical significance (P ≤ 0.01).

Key words: *grassland, fertilization, soil properties, respirometric activity*

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Activity of peroxidase in antioxidant protection of the cotton under salt stress

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Plants are greatly affected by salinity, which causes alteration in nutrient uptake, accumulation of toxic ions, osmotic stress, and oxidative stress. Salt stress induces the production of reactive oxygen species (ROS), and the response of peroxidase genes to this condition is tissue and developmental stage regulated. Peroxidases, as the key antioxidant enzymes, are widely distributed in nature and catalyze oxidation of various electron donor substrates concomitant with the decomposition of H_2O_2. In the management of the molecular and biochemical potential of the plant genotype using biologically active substances, DAG-1 (a supramolecular complex of glycerrhizin and salicylic acid) can be used to increase the tolerance of plants to the stress factors. The work was initiated to determine effect of various concentrations of DAG-1 (10^{-6}, 10^{-7} and 10^{-8} M) on increase of cotton species, Porloq-1 and Porloq-4, resistance to stresses. Each species is known to have specific biochemical parameters, so we need to determine a concentration in accordance with the parameters. Cotton seeds of Porloq-1 and Porloq-4 were the objects of the study. They were processed with the various concentrations of DAG-1 for 8 hours to be wrapped up in the paper rolls and placed in the separate vessels containing 100 mM of NaCl and water as a control. The seeds were germinated for 7 days in the thermostat at 27 °C. Method of Boyarkina was used to determine peroxidase activity spectrophotometrically. Our findings demonstrated that peroxidase activity for Porloq-1 species not processed with DAG-1 was 135.96 U/mg of protein and 137.52 U/mg of protein under the effect of salinization. In the sprouts of porloq-1, processed with DAG-1 in the concentrations of 10^{-6} peroxidase activity was found to increase by 44.1% in control experiments (without salinization) and accordingly 21.2% in salt stress (100 mM NaCl) Peroxidase activity in the seeds processed with DAG-1 in the concentrations of 10^{-7} and 10^{-8} remained as in the control. Porloq-4 was the cotton species to establish that peroxidase activity of unprocessed samples was 105.61 U/mg of protein. Processed with NaCl (100mM) peroxidase activity was 112.92 U/mg of protein. Peroxidase activity of seeds processed with DAG-1 in the concentrations of 10^{-6} and 10^{-7} remained as in the control. In the sprouts of Porloq-4 processed with DAG-1 in the concentration of 10^{-8} peroxidase activity increased by 30.65% in control experiments and accordingly 23.24% under salinity. Our findings demonstrated that DAG-1 not only regulates activity of peroxidase. Increase in the enzyme activity under salinity facilitates ROS detoxication. The DAG-1 concentration of 10^{-6} M was found to be the most efficient for the pre-sowing procession of cotton seeds of Porloq-1, while 10^{-8} M was the most suitable for Porloq-4.

Key words: *peroxidases, DAG-1, salt stress, reactive oxygen species, cotton*

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SECTION

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

Slaughter performance and meat quality of Holstein veal fed with total mixed ration (TMR) and alfalfa hay

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Supervisor: doc. Ing. Klára Vavrišínová, CSc.

The aim of this experiment was to investigate fattening, slaughter and quality parameters of Holstein calves fed with untreated total mixed ration (TMR) and alfalfa hay. Experiment included 20 male calves of Holstein divided into two groups according to feeding. Calves were during experimental phase housed in the same conditions – in group igloos, ten pieces each. Calves of the control group were fed with total mixed ration (TMR). Second group – experimental received industrially dried lucerne hay. After reaching the required slaughter weight, calves were after 193 days slaughtered in the experimental abattoir of the Department of Animal Husbandry, SUA in Nitra, dressed and chilled for 24 hours. Subsequently, detailed dissection of the right side of each carcass was performed. For the proximate composition, physical technological and sensory properties were samples of the loin (*M. longissimus lumborum et thoracis*) and top round (*M. semimembranosus*) muscles taken 24 hours after slaughter. Energy value in 100 g of meat sample was calculated according to the basic equation for calculating the energy value of the food. Meat acidity (pH) was measured in samples 1 hour and 24 hours *post mortem*. The individual evaluating of the veal quality were determined using laboratory techniques of SUA in Nitra. The experimental group of calves had higher average daily gains than control group (P > 0.05). No significant differences in the carcass weight were revealed (P > 0.05). Bulls of the control group had higher dressing percentage than those of experimental group (50.23%; P > 0.05). Proportion of kidney and intestinal fat was influenced by feeding concept; control group of calves had lower proportion of kidney fat (P > 0.05) and higher proportion of intestinal fat (P > 0.05). Differences in weights and proportions of individual carcass quarters were not significant (P.0.05). Also, differences in total amount of meat from right – half carcass were minimal (P.0.05). Proportion of trimmed fat was higher in the second group of calves (P.0.05). In terms of individual retail meat cuts, no significant differences between feeding groups were revealed (P > 0.05). Higher proportion of tenderloin was found in second group of calves (1.80%; P > 0.05). Statistically significant variety of the moisture content, intramuscular fat content and energy value as well, were revealed (P < 0.05). Physical technological parameters of both the muscles (pH, drip loss, electrical conductivity) showed similarity among the two feeding groups. Significantly higher electrical conductivity measured 24h post mortem were found in the second group (P < 0.01). Lightness (CIE L*) of the loin muscle after 24 hours *post mortem* was higher in the second group (P > 0.05). In colour spectrum of *M. longissimus thoracis* measured 7 days after slaughter we observed lighter (L* 47.17; P > 0.05) and pinker (a* 7.33; P > 0.05) meat in group fed with alfalfa hay. Colour parameters of top round muscle were similar in both groups (P > 0.05).

Key words: *Holstein, quality properties*, *proportion of tissues*, *meat quality class, slaughter value*

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Preparation and characterization of low molecular weight chitosans

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Chitosan is a cationic polysaccharide consisting of β-(1,4)-linked D-glucosamine and *N*-acetyl-D-glucosamine units. Chitosan is usually prepared by the deacetylation of chitin, the second most abundant renewable biopolymer in the world, which is mainly structured by β-(1,4)-linked *N*-acetyl-D-glucosamine residues. The molecular weight (MW) of chitosan ranges from 300 kDa to over 1000 kDa depending on the source and preparation procedure. High MW induced water insolubility and viscosity of chitosan limit its biological applications. Low molecular weight (LMW) chitosans are complete water-soluble molecules possessing a wide range of biological activities, which include antiviral, antibacterial, antifungal, antioxidant, anti-tumor, immune-enhancing, and lipid-lowering effects. Therefore, they have numerous promising applications in multiple fields such as medicine, cosmetics, food, and agriculture. The biological properties of LMW-chitosans tightly depend on their chain length. LMW-chitosans with variable degree of polymerizations (DPs) can be prepared via different approaches such as irradiation, acidic, enzymatic or oxidative depolymerization.

In this study, a chitosan sample (M_w = 150 kDa, DD = 85%) was chemically depolymerized with hydrogen peroxide in the presence of copper (II) ions. The reactions were carried out by applying various temperatures (10 – 60 ºC), time durations (10 – 120 min), concentration of hydrogen peroxide (1.0 – 9.0%), and amount of copper (II) acetate (16 – 160 mg). The depolymerized samples were freeze-dried following purification by dialysis against distilled water. The obtained LMW-chitosans were further studied with respect to molecular mass characteristics and structures through size-exclusion chromatography (SEC), IR- and NMR spectroscopic methods. In the course of the studies, LMW-chitosan samples (M_w = 2800 – 9200 Da, DP = 17 – 55) were prepared with a yield of 34 – 68%. The SEC results showed the samples obtained possess lower polydispersity index (1.10 – 1.31) than that of the starting material (2.25). The MW and product yield was decreased with the temperature, time and the peroxide concentration. The IR- and ^{13}C NMR spectroscopic studies showed the initial structure of the chitosan retains and side-reaction such as ring-opening, the dehydration neither occurs in the reaction, indicating that the polysaccharide undergoes the degradation by glycosidic bonds selectively. In conclusion, the depolymerization method can be used for preparation of completely water-soluble LMW chitosans. The LMW chitosans prepared are suitable for use in further studies or for applications in biomedical and agricultural purposes.

Keywords: *chitosan, depolymerization, low molecular weight chitosans, structure*

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Comparative study of lipid-binding properties of chitosan, alginate, and pectin

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Supervisor: Academician, Prof. Dr. Shavkat Salikhov

For the correction of gastrointestinal disorders developing in acute diarrheal infectious diseases, eubiotics, enzyme preparations, antispasmodics and a number of other groups of drugs are used in clinical practice, among which enterosorbents are mostly being of interest in recent years. Enterosorbents are substances that have a high sorption capacity, are not destroyed in the gastrointestinal tract, effectively binding and excreting from organism endogenous and exogenous toxic compounds, supramolecular structures and cells, used for the purpose of treatment and disease prevention. The most important medical requirements for modern enterosorbents are the high sorption capacity in relation to the removed components and the ability to sorb molecules and bacterial cells of different sizes and weights. Recent studies revealed naturally occurring polysaccharides are potential candidates to use as enterosorbents regarding their low toxic and highly sorption capacity.

In the present study, a comparative lipid-binding property was explored for chitosan obtained by the deacetylation reaction from chitin isolated from the *Artemia* cysts of the Aral Sea. Commercial pectin (pectolate, symbiotic based on apple pectin) and calcium alginate were used as comparing preparations. The lipid-binding ability of the dietary fibers (chitosan, pectin, and alginate) was studied with creating conditions that imitate the digestion of food in the stomach and intestine *in vitro*. Lipid-binding ability of the samples was calculated according to the following formula: $EL = (m_1 - m_0)/m_0$, where EL (g/g) – the value of lipid-binding ability, m_0 (g) - mass of the initial dry sample, m_1 (g) - mass of experimented sample, precipitate after centrifugation and drying. The results obtained for the comparative study of the lipid-binding abilities of the natural sorbents were shown in the following table.

Sample	Triglycerides (g/g)		Fatty acids (g/g)	
	$M \pm s$	Allowed interval	$M \pm s$	Allowed interval
Pectin	4.61 ± 0.09	$4.49 - 4.69$	3.22 ± 0.04	$3.17 - 3.27$
Calcium alginate	4.16 ± 0.05	$4.10 - 4.21$	3.17 ± 0.03	$3.13 - 3.21$
Chitosan	4.43 ± 0.59	$3.35 - 4.75$	3.21 ± 0.06	$3.15 - 3.29$

In the results, the samples under study showed lipid-binding properties with respect to triglycerides and fatty acids. The lipid-binding property of the chitosan prepared from *Artemia* cysts was found to be higher and lower than calcium alginate and pectin, respectively, regarding both for triglycerides and fatty acids (see table). In conclusion, chitosan prepared from Artemia cysts reduces the level of total cholesterol and low-density lipoproteins and so it can prevent the development of cardiovascular diseases such as dyslipidemia, hyperlipidemia, and atherosclerosis. Therefore, the chitosan possesses a perspective as a lipid-lowering additive to use in the food industry.

Keywords: *chitosan, preparation, Artemia cysts, lipid-binding property*

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SECTION

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

Extraction of seed oils with rich sources of fatty acids

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Development of flexible processes, green technologies using the knowledge in the basics of process engineering and employing mathematical modelling, optimization methods, computer-aided simulation, and exact results of experimental studies give an opportunity of real improvement of food and bio processing fields. Nowadays, researchers are performing to study of the effect of essential fatty acids on human health. Some of the plant seeds and by-products include oil with rich source of "essential" fatty acids. Firstly, to define the highest percentage of essential fatty acids and their ratio Soxhlet extraction method was applied. Result of the extraction and fatty acids profile showed that flax seed, rape seed and dog rose seed can be selected for future researches. Among them dog rose seed oil extraction processes have been studied by many scholars of the world but there is no data about waste of the dog rose in Uzbek variety. The main purpose of our research is to determine the best conditions of the process of extraction for high oil yield in lab conditions. Experiments carried out by the following methodology milled dog rose seed particles ($0.3 < d_1 < 1$ mm, $1 < d_2 < 2$ mm) balanced for each example 40 g then loaded in Erlenmeyer flask with 250 mL capacity, an amount of the solvent in ratios (solvent: seed ratio of 3 : 1 ml/g, 5 : 1 ml/g), the temperature range of the solvent (35 – 40 °C, 55 – 60 °C) were followed by design of experiment respectively. The results of regression analyses and optimization showed that highest oil was obtained at the highest process temperature (55 – 60 °C) and seed: solvent ratio (5 ÷ 1) and at the lowest particle size (0.3 ÷ 1) (studied ranges). Interaction among variables was not observed. Extracted oil was the highest in laboratory conditions, in the future should be studied on pilot scales as well as new technologies such as SFE, screw pressure and so on. Comparison of the technologies should be done.

Key words: *dog rose seed, EFA, PUFA*

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Mathematical modeling and optimization of the parameters of the process of extraction of water soluble polysaccharides from fruits *Prunus domestica* L.

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Supervisor: Sagdullaev Bakhodir Takhirovich, DSc.

The purpose of this work is to create a mathematical model and optimize the extraction process to identify the optimal conditions for isolating the water-soluble polysaccharides of *Prunus domestica* L. plum fruit necessary to create an original laxative based on them. Studies were carried out on the basis of single-factor experiments. They were conducted to collect a priori information, i.e. in each experiment, the parameters of only one of the factors influencing the process were changed, and the rest were left unchanged. However, it is known that the extraction process depends on many factors, each of which to a greater or lesser extent affects the productivity of the process. Assessing the overall situation is a very complex task and requires a large number of experiments to determine the boundary conditions and some parameters. To solve the problem of finding the optimal parameters of the extraction process, we decided to obtain a model of the process under study and optimize it using the Box-Wilson steep ascent method. Taking into account the results of single-factor experiments, the main factors affecting the extraction process were chosen: X_1 – the degree of grinding of raw materials; X_2 – hydronic module; X_3 – the speed of rotation of the centrifugal shaft; X_4 – filtration time. The levels of factors and the intervals of their variation were selected on the basis of a priori information. Due to the fact that a full factorial experiment would require setting $24 = 16$ experiments, taking into account repeated experiments $16*2 = 32$ experiments, to optimize the extraction process, we used ½ replicas of the full factorial experiment of type $Y = 24-1$, given to the generating relations: $X_4 = X_1*X_2*X_3$. Each of the 8 experiments was carried out in accordance with the compiled matrix, using the levels of each factor encoded in the matrix with + or - signs (respectively, the upper or lower level of variation). For example, in experiment 1, extraction was carried out with the plum fruits of domestic *Prunus domestica* L. with a grinding degree of 2 mm at a water ratio of 1: 5 and a centrifugal shaft rotation speed of 60 rpm for 2 hours. Thus, factors X_0, X_1, X_2, X_3 turned out to be significant factors, which is quite understandable. Increasing the temperature, the tangential velocity of the solution in the module and reducing the degree of concentration of the extract, naturally, should increase Y. Factors X_4 - are insignificant only in the selected interval of variation, and not in general during the filtration process. Statistical analysis of the data revealed that the mathematical model is adequate. Comparison of the confidence interval with the regression coefficients of the equation showed that the main factors affecting the process are: extraction temperature, hydraulic module, rotation speed of the centrifugal shaft, extraction time. According to the degree of their influence on the process, they are located in the following row: $X_1 > X_2 > X_3 > X_4$. Carrying out the process optimization made it possible to increase the yield of water-soluble polysaccharides to 47.6% of their content in the raw materials at the first contact of the phases, which was confirmed by a series of balance experiments.

The study of the antimicrobial activity of pectic substances derived from the fruit of *Prunus domestica* L.

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Supervisor: Sagdullaev Bakhodir Takhirovich, Dsc.

The widespread use of pectins in the food, medical and pharmaceutical industries makes it necessary to evaluate the biological effects of this group of compounds. These effects undoubtedly include the antimicrobial activity of pectic substances. One of the sources for obtaining pectic substances in the Central Asian region may be fruit and berry raw materials, such as fruits of the plant *Prunus domestica* L. In connection with the foregoing, the purpose of this work was to study the antimicrobial activity of pectic substances obtained from the fruits of the plant *Prunus domestica* L., which grows in Uzbekistan.

To determine the antimicrobial potential of pectic substances obtained from the fruit of *Prunus domestica* L., test bacteria from the collection of microorganism cultures of the Institute of microbiology of the Academy of Sciences of Uzbekistan (*Bacillus subtilis* 5, *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Candida albicans*) were used. The effect of substances on non-spore test cultures was determined in the exponential growth phase (after 36 – 42 h), and in spore cultures – at the stage of sporulation (after 48 – 72 hours). Evaluation of antagonistic activity was carried out on the 3 – 5th day of incubation according to the diameter of the sterile zones in the bacterial lawn formed around the wells.

The control was «Rutan», developed at the Institute of bioorganic chemistry named after Academician A. S. Sadykov of the Academy of Sciences of Uzbekistan based on the plant *Rhus coriaria*. The experiments were conducted in 2 replications.

The study of the antimicrobial activity of these plant substances in relation to the test cultures showed definite antagonistic and bacteriostatic activity. As a result of the research, it was established that pectic substances are capable of inhibiting the growth and development of conditionally pathogenic microorganisms (the zone of inhibition of the growth of the pathogen was 10 – 12 mm). At the same time, in relation to *Pseudomonas aeruginosa*, the lysis zone was 10 mm, to *Staphylococcus aureus* – 12 mm. And in relation to the test cultures of *Bacillus subtilis* 5, *Escherichia coli*, *Candida albicans*, the lysis zone was not observed. Thus, as a result of the research, it was established that pectic substances from the fruit of *Prunus domestica* L. can significantly inhibit the growth and development of conditionally pathogenic cultures.

Key words: *pectins, Prunus domestica L., antimicrobial activity*

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Utilizing the technology of targeted must oxygenation in the process of producing still white wines in order to reduce the need for the application of sulfur dioxide and to enhance the varietal character of the wine

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The aim of the experiment was to physicochemically and sensorically compare effects of targeted must 's oxygenation and conventional technology in the pre-fermentation process of wine production on phenolic compounds, the need for sulfur dioxide application and sensory expression of wines. We used the world-wide white wine grape variety Chardonnay. The principle of the oxygenation method is to oxidize phenolic substances. They are responsible for the oxidation processes in the wine and their removal during the clarification process in the grapes processing process. In the method of targeted oxygenation of must, preparations containing sulfur dioxide and antioxidant preparations are not applied during the processing of the grapes until the clarification. In the control variant, sulfur dioxide was applied during the processing of the grapes. In the conventional method, sulfur dioxide and antioxidant preparations are already used during the processing of grapes into mash and unscale must.

Total phenol content was determined by spectrophotometry at 765 nm. We carried out the evaluation of organoleptic characteristics of wines by means of a hundred-point evaluation system according to the International Organization of Vine and Wine (OIV) and the International Union of Oenologists (UIOE). The amount of total sulfur dioxide was determined by the iodometric titration method. We used the method of analysis of variation in Statgraphics Centurion XVII (StatPoint Inc. USA) to statistically evaluate the results.

The total content of phenolic substances in wine produced without the use of targeted oxygenation of must was 185 mg/l. By the method of targeted oxygenation their total content was statistically demonstrably lower, namely 152 mg/l. The variant produced by targeted oxygenation contained statistically significantly less phenolic substances, which in white wine are the oxidizing medium responsible for premature aging of the product.

The wine produced by targeted oxygenation contained 78 mg/l of total sulfur dioxide. The sample produced by the conventional method contained up to 122 mg/l of total sulfur dioxide, which represents a 36.1% difference against the control variant.

Sensory evaluation took place in two terms. The first evaluation took place 30 days after the wine was bottled. The oxygenated variant scored 85.4 points and the sample manufactured conventionally scored 86.6 points. We did a sensory evaluation again after 12 months of storing wine samples under standard conditions. The oxygenated variant scored 85 points and the conventionally produced sample was 82.5 points.

The results obtained show that the method of targeted oxygenation makes it possible to limit the need for the use of sulfur dioxide in the oenological process for the production of white still wines without adversely affecting the ripening processes, the sensory profile and the varietal character of the wine.

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