

Innovative and Applied Research in Biology Inovatīvie un Pielietojamie Pētījumi Bioloģijā



Programme and abstracts

2024 Riga





Innovative and Applied Research in Biology

Inovatīvie un Pielietojamie Pētījumi Bioloģijā

Friday, 8 March 2024, 10.00 AM, ZOOM Jelgavas street 1, 702.

Programma / Programme

| 10.00–10.05 | Līga Jankevica Institute of Biology, University of Latvia | Opening Atklāšana | | |
|---|--|--|--|--|
| Vadītājs/Chair: Prof. Dr. hab. biol. Īzaks Rašals | | | | |
| O1 10.05–10.35 | Algimantas Paulauskas Vytautas Magnus University | Colaboration between geneticists from the Baltic countries in Conservation genetics and Molecular Ecology | | |
| O2 10.35–10.50 | leva Ignatavičienė Nature Research Centre | Genetic diversity and variation of ten duckweed (L. minor) populations based on ascorbate peroxidase gene and microsatellite markers | | |
| O3 10.50–11.05 | Dace Grauda LU Institute of Biology | Impact of nanoparticles and microparticles (succinite, Ag, SiO2, Al2O3) on plant gametic cells | | |
| O4 11.05–11.20 | Gabija Steigvilaitė Kaunas University of Technology | Sustainable processing of secondary plant-based raw materials using biological methods | | |
| O5 11.20–11.35 | Laura Āboliņa LU Institute of Biology | Cloudberry cultivation in temperate climates: insights from blueberry, cranberry, and lingonberry studies in peat fields | | |
| 11.35–12.00 | Coffee break / Kafijas pauze | | | |
| Vadītājs/Chair: Dr. biol. Prof. Dalius Butkauskas | | | | |
| O6 12.00–12.15 | Rolands Krams Daugavpils University | Permanent feeders decrease the survival of wintering great tits (<i>Parus major</i> Linnaeus, 1758) | | |
| O7 12.15–12.30 | Antra Stīpniece Lab. of Ornithology, LU Institute of Biology | Marking Greylag Goose <i>Anser anser</i> with colour neckbands in Latvia - the first results. | | |
| O8 12.30–12.45 | Oskars Keišs Lab. of Ornithology, LU Institute of Biology | Monitoring of autumn migration of birds at Pape, Latvia 1992–2023 | | |
| O9 12.45–13.00 | Viesturs Vigants Lab. of Ornithology, LU Institute of Biology | A novel approach for tracing migration patterns of a short-distance migrant, the common starling <i>Sturnus vulgaris</i> | | |

| | Evelina Juozaitytė- Ngugu | Overview of Sarcocystis species parasitizing birds |
|-----------------|---|--|
| | Nature Research Centre | |
| 13:15 – 14:15 | Lunchbreak / Pusdienu pārtraukums | |
| Vadītājs/Chair: | Dr. hab. biol. Īzaks Rašals | |
| | Stenda referātu prezentācijas (2 min), jautājumi (1 min) Short poster presentations (2 min), Questions (1 min) | |
| | Gabriela Liubartaitė Nature Research Centre | Microsatellite Genotyping of <i>Cyprinus carpio</i> Czech and Lithuanian Strains |
| | Evelina Maziliauskaitė Nature Research Centre | Molecular Investigation of Sarcocystis spp. in Invasive Raccoon (<i>Procyon lotor</i>) from Lithuania |
| | Viktorija Levinger Nature Research Centre | Intraspecific genetic variability of Cloudberry (<i>Rubus chamaemorus</i>) in the Eastern Baltic region based on microsatellite data analysis |
| | Nadežda Krasņika LU Institute of Biology | Optimization of oat (<i>Avena sativa</i> L.) embryo culture method for breeding |
| | Edīte Juceviča Institute of Biology, LU; University of Life Science and Technologies | Preliminary results of soil mesofauna characterized by QBS index in the background of restoration success of urban grasslands in Riga city, Latvia |
| | Irina Sivicka Latvia University of Life Sciences and Technologies | Effect of treatments with microalgae extracts to total chlorophyll content in tomato leaves |
| | Kristaps Neiberts LU Institute of Biology | Mixotrophic cultivation of microalgae in whey medium for poultry diet supplementation with microalgal biomass |
| | Svetlana Vasiljeva, LU Institute of Biology | Optimization of the production of sodium alginate derivatives |
| | Abbas Mirshafiey Int.Reviewer/FISM (Italian Multiple Sclerosis Society Foundation) | The anti-inflammatory effect of Mannuronic acid (M2000) in experimental model of glomerulosclerosis |
| | Abbas Mirshafiey Int.Reviewer/FISM | Anti-tumoral effects of Guluronic acid (G2013), as a novel safe natural agent on animal model of breast cancer |
| | Abbas Mirshafiey Int.Reviewer/FISM | Inhibitory property of Mannuronic acid on angiogenesis phenomenon using experimental model |
| | Kārlis Žagata LU Institute of Biology | Different model systems to evaluate EMR impact |
| 15:00-15:30 | Coffee / Kafijas pauze | |

| Vadītājs/Chair: Dr. biol. Prof. Dalius Butkauskas | | | |
|---|---|---|--|
| O11 15:30- 15:45 | Ilva Trapiņa LU Institute of Biology | Report on the project's second year "Development of an innovative approach to identify biological determinants involved in the between-animal variation in feed efficiency in sheep farming." | |
| O12 15:45 - 16:00 | Samanta Pļaviņa LU Institute of Biology | The feed efficiency of intensive fattened lambs in Latvian sheep breeds is linked to genetic variations of the MSTN gene. | |
| O13 16:00 - 16:15 | Tatjana Grigorjeva Daugavpils University | Impact of serotonin concentration changes on the laterality of fruit flies (<i>Drosophila melanogaster</i> Meigen, 1830) | |
| O14 16:15 - 16:30 | Ineta Salmane LU Institute of Biology | Fauna of the predatory Mesostigmata mites (Acari, Parasitiformes) in the urban grasslands of Rīga city | |
| 16:30 -17 :00 | Noslēgums, diskusijas Conclusions, discussions | | |

Innovative and Applied Research in Biology

Abstracts

Oral Presentations

Colaboration between geneticists from the Baltic countries in Conservation genetics and Molecular Ecology

Algimantas Paulauskas Vytautas Magnus University, K. Donelaičio str. 58, 44248, Kaunas Corresponding author e-mail algimantas.paulauskas@vdu.lt

The field of conservation genetics and molecular ecology has witnessed significant growth and advancements, primary among these is that molecular ecology itself revolves around the way genetics and species development are affected by ecological and environmental factors. Geneticists from Baltic countries gather genetic data on individuals and populations of species of conservation concern and use this information to address a wide variety of questions. Their Work on questions related to molecular evolution, behavioral ecology, population dynamics and gene flow, the ecology of infectious disease, and the detection of invasive species. Here, we present selected aspects of collaboration in population genetics, genome evolution, and molecular phylogeny with an emphasis on contributions by Baltic countries researchers.

Key words: Population genetics, genome evolution, molecular phylogeny, Baltic countries

Genetic diversity and variation of ten duckweed (*Lemna minor*) populations based on ascorbate peroxidase gene and microsatellite markers

Ieva Ignatavičienė¹, Dalius Butkauskas¹
¹Nature Research Centre, Akademijos Street 2, Vilnius 08412, Lithuania Corresponding authors e-mail: ieva.ignataviciene@gamtc.lt

Lemna minor is known as common duckweed, which is a small, free-floating, and fast-growing aquatic plant. The species is distributed worldwide and often grows on the surface of still or slow-moving, nutrient-rich, and fresh waters. Analyzing duckweed genetic diversity is essential for germplasm preservation, academic studies, and various practical applications.

L. minor specimens from ten locations were collected from different water courses from small ponds to lakes and rivers in Lithuania and Latvia. Their genetic variability was assessed based on sequences of the ascorbate peroxidase (APx) gene (A1) and two chloroplast microsatellites (L4, L14). The largest number 44 of unique haplotypes were identified among the populations, based on APx gene sequence analysis. 25 and 31 unique haplotypes were obtained using L4 and L14 microsatellite markers, respectively. Based on Tajima's D statistic test, we determined that the collected ten populations of duckweed were stable. Analysis of molecular variance (AMOVA) results indicated that genetic variations occurred among populations, ranging from 5% (L4) to 27% (L14). Haplotype network, PCoA, and phylogenetic analyses demonstrated apparent genetic diversification between VN, V (Neris River) and LV, L (Daugava River) population groups. The results of Mantel tests showed that the genetic distance among geographical populations of L. minor was positively correlated with the geographical distance and the genetic distances using A1 and L4 markers ($R^2 = 0.437$, P=0.04 and $R^2=0.4104$, P=0.04, respectively), which indicates that geographical isolation is an important factor for the observed genetic differentiation.

Our results showed different informative markers for population genetic studies in *Lemna minor*. The identified genetic diversity and population structure of duckweed expand our understanding of the genetic background of this species.

Key words: Lemna minor, genetic diversity, chloroplast microsatellites, ascorbate peroxidase

Impact of nanoparticles and microparticles (succinite, Ag, SiO₂, Al₂O₃) on plant gametic cells

Dace Grauda¹, Alise Aglinska¹, Dalius Butkauskas²
¹University of Latvia, LV-1004 Rīga, Jelgavas Street 1, Latvia; ²Nature Research Centre, LT-08412 Vilnius, Akademijos Street 2, Lithuania Corresponding authors e-mail: dace.grauda@lu.lv

Since the progress of nanotechnology nanoparticles as well as microparticles have been studied in various aspects, including environmental impact and human health. Nanoparticles depending on their size can penetrate in cell through cell membrane, accumulate there, and affect molecular mechanisms in cell, microparticles whose size does not allow them to enter the cell can stick to the cell surface membrane and through it affect cell transport pathways.

In this research the impact of four types of nano- and microparticles: SiO2 (0.2-0.3 μm), Al2O3 (2.5 μm), Ag (0.5-1 μm) and succinite (5 nm - 3 μm) on immature Hordeum vulgare microspores were studied. The reason for choosing these particles is because of their frequent occurrence in everyday life (cosmetics, paints, textile etc.). The immature microspore culture was used to reveal the effects of microparticles because it has already been identified as a very sensitive system for bioindication. Genetically identical microspores were incubated in vitro for 1.5 hours in cultures containing nanoor microparticles. Relative cell autofluorescence was measured with BD FACSJazz® cell sorter. DNA was extracted from cells incubated with nano- and microparticles, and iPBS fingerprinting method with various primers was utilized to observe if retrotransposon changes occur in samples incubated in media with added different particles. In general, Al₂O₃ and SiO₂ microparticles have a suppressing negative effect on the autofluorescence of microspores, while succinite (amber) particles increase it, and Ag particles have no effect on fluorescence of microspores in this study. The iPBS method revealed changes in the spectrum of amplified barley microspore DNA in samples incubated with SiO₂, Al₂O³ and succinite nano- and microparticles.

Acknowledgments: The research is supported by project Nr. ES RTD/2022/7 "3D Biotextile with Technological Composition of nano particles to enhance the protecting properties" (Latvia) and project Nr. S-M-ERA-NET-22-1 "3D Biotextile with Technological Composition of nano particles to enhance the protecting properties" (Lithuania)

Key words: barley microspores, bioindication, impact on autofluorescense, retrotransposon changes

Sustainable processing of secondary plant-based raw materials using biological methods

Authors: Gabija Steigvilaitė, Lina Vaičiulytė

Affilliation: Faculty of Chemical Technology, Kaunas University of Technology,

Radvilėnų ave. 19, LT-50254, Kaunas, Lithuania

Corresponding author e-mail:: gabija.steigvilaite@ktu.edu.lt

During the processing of fruits, berries and other plant products, large amounts of pomace are formed, containing many valuable organic and inorganic substances. In order to achieve sustainable agricultural development and organize food production according to the principles of sustainable technologies, it is important to explore effective ways of utilizing and adapting plant pomace for the creation of new products.

The scientific literature and industrial practices present various traditional chemical and physical methods for the processing of plant pomace, which have certain limitations in terms of energy and solvent requirements [1]. Increasing attention is being given to eco-friendly methods of processing plant waste to reduce the use of the mentioned resources. One way of valorizing plant waste is through biological processing, such as fermentation.

Lactic acid bacteria (LAB) fermentation is a popular method, but fermentation with LAB alone may not be effective. It is particularly relevant to explore spontaneous fermentation (e.g., *Medusomyces gisevii*) and to combine it with traditional LAB fermentation. The addition of LAB during fermentation not only inhibits the growth of harmful microorganisms and ensures the preservation of the final product, but also enhances its probiotic properties.

The aim of this work is to apply spontaneous (*Medusomyces gisevii*) and cultural (LAB) fermentation methods for sustainable bioprocessing of plant pomace and to apply the selected biological matrices for the production of plant-based beverages.

The composition of the natural symbiotic consortium Medusomyces gisevii is diverse and dependent on the conditions, place, and time of cultivation. Therefore, genetic studies of this culture will be conducted during the research for identification purposes. Medusomyces gisevii was used for the production of a fermented beverage from raspberry pomace. Additionally, homofermentative L. plantarum heterofermentative *L. reuteri* cultures were used in the fermentation process, which produce bacteriocins and organic acids that protect against technologically harmful microorganisms. The resulting beverages were characterized by nutritionally significant LAB concentrations, good sensory properties and increased consumer acceptability. The results of the study show that raspberry pomace can be used for the production of a plant-based probiotic beverage.

Key words: plant by-products, bioprocessing, fermentation, symbiotic culture, lactic acid bacteria

References

1. Waseem, M., et al. Conventional and advanced extraction methods of some bioactive compounds with health benefits of food and plant waste: A comprehensive review. Food Frontiers, 4, 1681–1701, 2023.

Cloudberry cultivation in temperate climates: insights from blueberry, cranberry, and lingonberry studies in peat fields

Laura Āboliņa ¹, Andis Karlsons¹

¹University of Latvia, Institute of Biology, Laboratory of Plant Mineral Nutrition, O. Vācieša street 4, Riga, Latvia

Corresponding authors e-mail: abolina.la@gmail.com

The cultivation of highbush blueberry and American cranberry has been extensively studied; however, concerning the mineral nutrition of plants, research predominantly covers nitrogen fertilization experiments. Lingonberry and cloudberry have been studied scarcely; nevertheless, in Latvia, interest is increasing in growing these berries. Thus far, cloudberry growing experiments have been conducted in polar climate regions; therefore, growing technology must be developed for the temperate climate zone. Firstly, the propagation and rooting of cloudberry seedlings are essential for healthy planting material. Once planted, cloudberries require optimal moisture, lighting, and fertilization management for successful plant development and high yields. Additionally, successful fertilization of berries in peat fields can be challenging due to high nutrient leaching rates.

In this review, we develop a plan for starting cloudberry cultivation in the climatic conditions of Latvia, based on knowledge about blueberry, cranberry, and lingonberry cultivation in peat fields. We assess the trends of plant mineral nutrition in berry plantations over the last 20 years from the literature and conclude that nutrients N, S, Cu, Mo, and B are most often deficient in peat fields. Based on a literature review and experience with fertilization plan development, we estimate the optimal nutrient levels and application rates for the new berry species. Current knowledge indicates cloudberry fertilization requirements are lower than those of highbush blueberry and American cranberry regarding the amount of fertilizer, yet similar regarding the type of fertilizer. Reviewing the most common problems in mineral nutrition and overall plantation management of acid peat berry species will help determine the first steps toward developing growing technology for cloudberry in Latvia.

Key words: Rubus chamaemorus, fertilization, mineral nutrition, acid peat, growing technology.

Permanent feeders decrease the survival of wintering great tits (*Parus major* Linnaeus, 1758)

Ronalds Krams^{1,2,3}, Tatjana Krama^{1,2,3}, Sergejs Popovs^{1,2}, Giedrius Trakimas^{1,4}, Markus J. Rantala⁵, Todd M. Freeberg^{6,7}, Māris Munkevics^{1,2,8}, Indriķis Krams^{1,2,8,9}
¹ Daugavpils University, Institute of Life Sciences and Technologies, Daugavpils, LV-5401, Latvia; ² Latvian Biomedical Research and Study Centre, Rīga, LV-1067, Latvia; ³ Estonian University of Life Sciences, Institute of Agricultural and Environmental Sciences, Tartu, 51006, Estonia; ⁴ Vilnius University, Institute of Biosciences, Vilnius, 10257, Lithuania; ⁵ University of Turku, Department of Biology, Turku, 20014, Finland; ⁶ University of Tennessee, Department of Psychology, Knoxville, TN 37996, USA; ⁷ University of Tennessee, Department of Ecology and Evolutionary Biology, Knoxville, TN 37996, USA; ⁸ University of Latvia, Faculty of Biology, Rīga, LV-1010, Latvia; ⁹ University of Tartu, Institute of Ecology and Earth Sciences, Tartu, 50409, Estonia. Corresponding authors e-mail: ronalds.krams@du.lv

Survival for birds can be challenging during the cold winter months in Northern Europe. Humans often provide food to wintering birds, thus, improving their winter survival. The optimal body mass hypothesis posits that the body reserves of wintering birds are balanced between the risk of starvation and predation.

In this study we tested whether the body mass of wintering great tits (*Parus major* Linnaeus, 1758) was higher under conditions of less predictable food resources. We compared body mass, body mass index, the speed at take-off and apparent survival of great tit adult males wintering in small urban areas, either near feeders providing permanent access to food for months, or near feeders providing irregular access to food. Body mass and body mass index were greater, while take-off speed and apparent survival were lower in birds wintering near permanent feeders than birds wintering near irregular feeders. Thus, urban birds, with their predictable access to high energy food, did not follow the fattening strategy predicted by the optimal body mass hypothesis.

This study shows that excess under skin fat reserves of great tits wintering near permanent feeders can make them slow at take-off, which increases their exposure to predators. Regular excess amounts of high-energy food may affect urban birds' physiological and behavioral strategies in a non-adaptive way.

We suggest that caution should be taken when choosing a proper place to position bird feeders to prevent making birds at feeders' easy prey for predators. We recommend irregular feeding of wintering birds and the placing of feeders in places that are safe against attacking predators.

Key words: *Parus major*, bird feeders, take-off speed, winter fattening, passerines, survival

Marking Greylag Goose *Anser anser* with colour neckbands in Latvia - the first results

Antra Stīpniece, Andris Stīpnieks, Jānis Bētiņš, Dmitrijs Boiko, Toms Endziņš, Oskars Keišs, Artūrs Laubergs.

Lab of Ornithology, Institute of Biology, University of Latvia Corresponding authors e-mail: antra.stipniece@lu.lv

Greylag Goose is a huntable species in Latvia (open season both in autumn and spring in early 70ties; then hunting forbidden; hunting resumed in 2004, but only in autumn). Its breeding population has risen to from 1 known pair in 1957 (Vīksne, Mednis 1978) to 500 pairs (Ķerus u.c. 2021). As for a scarce breeder, the ringing efforts have been low and before 2021 no recoveries of Greylag Geese ringed in Latvia are available.

In 2021 and 2023 19 flightless Greylag Geese were trapped and marked with metal leg bands and yellow plastic neck bands. Five individuals (4 juveniles and an adult female provide resighting history outside the marking place. Most of the resightings come from south-west Lithuania wetlands where Greylag Geese hatched in Engure Lake arrived already in August-September. Two resightings (September, December) come from Poland, three winter resightings (October, November) from Germany, reaching maximum distance 741 km from the ringing place. The possible reasons of the migration distance and direction observed are discussed. Next summer 2 juveniles returned to the hatching lake, but for one bird all resightings up till August occurred in Lithuania.

Acknowledgments. The work was possible due to funding by Vides aizsardzības fonds and Medību saimniecības attīstības fonds. We thank all colleagues who helped with geese capture.

Key words: staging, wintering, juvenile dispersal

Monitoring of autumn migration of birds at Pape, Latvia 1992–2023

Oskars Keišs, Ivo Dinsbergs, Māris Jaunzemis, Valts Jaunzemis Laboratory of Ornithology, Institute of Biology, University of Latvia Corresponding authors e-mail: oskars.keiss@lu.lv

Monitoring of migrating passerine birds by standartized capture in Heligoland type trap at Pape, at the Latvian Coast of the Baltic See has been carried out since 1992. However birds in Pape have been captured since 1966 (Blums et al. 1967). First visual observations have been done in 1958 (Michelsons u.a. 1960). The full analyses of all of the collected data are not possible yet, since old data are still available only on paper.

During the period of 33 autumn seasons (1992-2023), three species show steep decline in numbers: Redwing (Turdus iliacus), Blue Tit (Parus caeruleus) and Blackcap (Sylvia atricapilla). Further 16 species show moderate decline: Sparrowhawk (Accipiter nisus), Lesser Whitethroat (Sylvia curruca), Chifchaff (Phylloscopus collybita), European Robin (Erithacus rubecula), Great Tit (Parus major), Garden Warbler (Sylvia borin), Wood Warbler (Phylloscopus sibilatrix), Wren (Troglodytes troglodytes), Crested Tit (Parus cristatus), Goldcrest (Regulus regulus), Bullfinch (Pyrrhula pyrrhula), Coal Tit (Parus ater), Willow Tit (Parus montanus), Treecreaper (Certhia familiaris) and Blackbird (Turdus merula). Eight species showed stable trend: Siskin (Carduelis spinus), Spotted Flycatcher (Muscicapa striata), Lesser Spotted Woodpecker (Dendrocopos minor), Pied Flycatcher (Ficedula hypoleuca), Brambling (Fringilla montifringilla), Song Thrush (Turdus philomelos), Great Spotted Woodpecker (Dendrocopos major), and Common Redstart (Phoenicurus phoenicurus). Only three species showed increasing trend during our 32 year study period: Firecrest (Regulus ignicapilla) Cuckoo (Cuculus canorus), and Chaffinch (Fringilla coelebs). Two irruptive species had no clear trend Nuthach (Sitta europaea), and Long-tailed Tit (Aegithalos caudatus).

Standartized visual observation data are available since 2017, it is much shorter period and the trends for some species are contradictory to those of capture data since 1992.

References:

Blūms, P., Baumanis, J., Baltvilks, J. 1967. Bird trapping with nets during the autumn migration 1966 in Latvia [in Latvian with English Summary]. *Zooloģijas muzeja biļetens* 1: 103–106. Michelsons H., Ģ. Kasparsons, G. Lejiņš, J. Vīksne, V. Šmits, J. Lipsbergs, I. Stolbovs. 1960. Die Migration der Vögel in der Lettischen SSR im Herbst 1958. [Ins Lettisch mit Deutscher Zusammenfassung] *Latvijas Putnu dzīve – Ornitoloģiskie pētījumi 2, Latvijas PSR Zinātnu*

akadēmijas Bioloģijas institūta raksti XIV: 139–192.

A novel approach for tracing migration patterns of a short-distance migrant, the common starling *Sturnus vulgaris*

Authors: Viesturs Vīgants¹, Ivo Dinsbergs¹, Māris Jaunzemis¹, Valts Jaunzemis¹, Ance Priedniece¹, Elza Zacmane¹, Oskars Keišs¹, Mārtiņš Briedis^{1,2}
Affiliation: ¹ Lab. of Ornithology, Institute of Biology, University of Latvia; ²Dep. of Bird Migration, Swiss Ornithological Institute, Switzerland Corresponding authors e-mail viesturs.vigants@lu.lv

Over the past decade, classical light-level geolocation has significantly advanced our understanding of bird migration. However, the inherent margin of error in determining geographic locations has impeded the effective tracking of short-distance migrants. This study addresses this limitation by integrating light-level geolocation with activity and barometric pressure tracking, offering a comprehensive analysis of the migration behavior of Common Starlings (*Sturnus vulgaris*) tagged during four breeding seasons (2020 – 2023) in Latvia.

To estimate the migratory patterns our study embraces the newly released R package GeoPressureR, which helps to incorporate geolocator data recordings with remotely sensed surface-level atmospheric pressure and wind data from The Copernicus Climate Change Service. Results demonstrate a remarkable correlation (r=0.99±0.01, p<0.001) between recorded and remotely sensed atmospheric pressure values at the breeding site, yielding location estimates with a maximum error of 30 km. Most tracked birds exhibited wintering patterns in the British Isles (~2000 km from the breeding site), with stop-over sites identified near the southern coast of the Baltic Sea and the Wadden Sea.

Two distinct migration strategies were observed among the Starlings: (1) approximately half of the birds initiated migration immediately after the breeding season in early June, engaging in three migration phases throughout the annual cycle (summer-autumn-spring); (2) the remaining individuals adhered to a classical autumn-spring migration behavior, remaining close to the breeding sites until late autumn. Intriguingly, contrary to existing literature, majority of long-distance migratory flights (~70%) occurred at night, commencing right after sunset, with one individual completing a significant portion of autumn migration in a single 22-hour flight. Notably, even in the absence of mountainous barriers, maximum flight altitude reached up to 2500 meters above sea level.

This study underscores the potential of multi-sensor tracking to provide unprecedented insights into the migratory behavior of individual birds throughout their entire annual cycle. Furthermore, it enhances the precision of geographical locations derived from light-level geolocators, showcasing the utility of this integrated approach in advancing our understanding of avian migration.

Acknowledgments: This study was funded by the Latvian Council of Science (project No. Izp-2019/1-0242 and No. Izp-2023/1-0233)

Key words: migration, geolocators, short-distance, atmospheric pressure, starling

Overview of Sarcocystis species parasitizing birds

Evelina Juozaitytė-Ngugu, Petras Prakas, Jolanta Stankevičiūtė, Rasa Vaitkevičiūtė, Dalius Butkauskas

Nature Research Centre, Akademijos 2, LT-08412 Vilnius, Lithuania Corresponding authors e-mail: juozaityt.evelina@gmail.com

Protozoan parasites of the genus *Sarcocystis* are cyst-forming coccidians widespread in mammals, birds and reptiles. These parasites are characterized by an obligatory heteroxenous two-host prey-predator life cycle. Asexual multiplication with formation of sarcocysts occurs in the intermediate host, while sexual stages develop in the small intestine of the definitive host. Birds may serve as intermediate hosts or definitive hosts for *Sarcocystis* spp.

Currently, birds are known to be intermediate hosts for 40 different species of Sarcocystis around the world, with 28 of these being validated. Only 15 Sarcocystis spp. parasitizing birds have known definitive hosts. Some Sarcocystis spp. found in birds complete their life cycles in specific intermediate hosts or within closely related host species, while other species appear less host specific. Several Sarcocystis species, S. calchasi, S. falcatula, S. halieti, and S. wobeseri, have been found in the muscles of different bird orders. Meanwhile, some Sarcocystis spp. are strictly host-specific.

It is important to note the observed pattern that *Sarcocystis* spp. with the lowest host specificity are pathogenic. The most pathogenic *Sarcocystis* species found in birds are *S. calchasi*, *S. falcatula* and *S. halieti*. These species can cause encephalitis, hepatitis and pneumonia. Acute infections with Sarcocystis parasites can also lead to the death of the birds.

Sarcocystis rileyi, found in the birds muscles of the order Anseriformes, cause infection, which might be considered as pathogenic to the host. Some ducks infected with this Sarcocystis sp. show signs of myopathy. Furthermore, severe infections of macrocysts in muscles of birds used for locomotion may cause a reduced flying capacity and ability to withstand migration, weakness, and an increased probability of becoming a victim of predators. Also, hunted birds infected with macrocysts of S. rileyi are not suitable for human consumption.

Birds have been shown to be definitive hosts of at least 22 *Sarcocystis* spp. worldwide. Based on phylogenetic studies, birds of prey and omnivorous birds are involved in transmitting even more *Sarcocystis* spp. that use carnivores, ungulates, birds and rodents as intermediate hosts.

Key words: Sarcocystis, S. rileyi, birds, intermediate and definitive host, parasites

Report on the project's second year "Development of an innovative approach to identify biological determinants involved in the between-animal variation in feed efficiency in sheep farming"

I.Trapina¹, D.Kairisa², N.Paramonova¹

¹ Genomics and Bioinformatics, Institute of Biology of the University of Latvia, Riga, Latvia; ² Department of Animal Sciences, Latvian University of Life Sciences and Technologies, Jelgava, Latvia;

Corresponding authors e-mail: ilva.trapina@lu.lv

Cooperation project (Izp-2021/1-0489) is interdisciplinary research related to economic activity in agriculture and biotechnology, implemented by the University of Latvia in collaboration with the Latvian University of Life Sciences and Technologies. This study aims to determine genetic and molecular markers to identify individual animals in sheep herds with the maximum predisposition to feed efficiency and weight gain, with the aim of their introduction into selection.

The project has completed genotyping DNA samples from the first collection, identifying 631 variable loci in 10 genes in 72 samples of six sheep breeds. The NGS method was used to genotype the entire region of five genes (UCP3, UCP2, NPY, GHRL, MSTN) and the exon region of five other genes (IGF2, IGF1, CAST, PPARG, and MTOR). The results were used in association analysis with feed efficiency indicators, such as Feed efficiency, Feed conversion ratio, Relative growth rate, Kleiber's ratio, Residual feed intake, Residual weight gain, and Residual intake and body weight gain.

The results from the association analysis for feed efficiency were used to select polymorphisms to be used to realize the goal of the project, i.e. to create a statistically reliable prediction algorithm for the selection of the Latvian dark-head and Latvian sheep breeds as a whole.

In parallel with the genotyping process, the fattening of lambs that were included in the second experimental group continued at the Klimpas intensive feeding station of the Latvian Sheep Breeders Association.

A total of 92 lambs underwent blood collection for DNA and RNA extraction, as well as biochemical studies, also information about phenotypic indicators were collected. During the second year of the project, the results of the first year of the project were presented at 11 presentations in international conferences, two scientific publications, and one popular scientific publication. Also, in cooperation with project partners, a publication on the possible economic significance of feed efficiency was prepared.

In 2024, it is planned to (1) create algorithm prototypes and test them in the second year's collection; (2) perform genotyping of selected, statistically associated polymorphisms in the second year's collection; (3) collect the phenotypic indicators of the second year's collection; (4) prepare results for publications; (5) to license the prepared algorithms as sciences at the University of Latvia.

Acknowledgments. The study was funded by the LZP-2021/1-0489 project: "Development of an innovative approach to identify biological determinants involved in the between-animal variation in feed efficiency in sheep farming".

Key words: sheep, feed efficiency, markers, DNA, prediction algorithm, polymorphisms

The feed efficiency of intensive fattened lambs in Latvian sheep breeds is linked to genetic variations of the MSTN gene

I.Trapina¹, S. Plavina¹, N. Krasņevska¹, J.Paramonovs¹, Z. Bardina¹, D.Kairisa², N.Paramonova¹

¹ Genomics and Bioinformatics, Institute of Biology of the University of Latvia, Riga, Latvia; ² Department of Animal Sciences, Latvian University of Life Sciences and Technologies, Jelgava, Latvia.

Corresponding authors e-mail: Samanta.plavina@lu.lv

Economic profitability in sheep farming directly depends on the level of feed efficiency of the lambs raised. Higher levels of feed efficiency directly correlate with increased economic benefits. Encouraging breed selection based on the feed efficiency tract allows sheep farmers to produce high-quality lambs while increasing economic profitability.

Marker association selection (MAS) is a selection method in livestock production that uses genetic biomarkers that have previously been associated with a particular trait. Potential feed efficiency MAS genes are responsible for the synthesis of proteins involved in food digestion and the development and enlargement of muscle and fat cells. Additionally, these genes are engaged in energy metabolism pathways. Myostatin (MSTN) is a well-preserved member of the transforming growth factor-beta superfamily. This protein plays a crucial role in controlling the process of muscle formation and functions as an inhibitor of muscle growth and development in animals. Various symptoms frequently arise from mutations occurring at different loci of the MSTN gene. The study aims to identify potential molecular markers associated with feed efficiency indicators in the MSTN gene for Latvian sheep breeds.

Genomic DNA was extracted from blood samples obtained from 76 controlled fatten lambs, of which 63.16% belonged to the Latvian dark-head (LT; Latvijas tumšgalve) breed; full sequencing of the MSTN gene, and analysis of loci variability was carried out. The identified variable loci were subjected to association analysis with feed efficiency indicators. The 6'757 bp length MSTN gene region has 204 polymorphism sites that are described in the Ensemble database; 23 polymorphic loci were identified, including one new SNP, information about which is not available in the Ensemble database. Three distinct complete linkage disequilibrium groups contained nine of the loci under investigation.

Significant statistical relationships were observed between SNP rs404916326 of the MSTN gene and Residual feed intake (RFI), Residual intake, and body weight increase (RIG) in the group of LT breed sheep. The single nucleotide polymorphism rs408469734 was discovered to be linked to both relative growth rate (RGR) and Kleiber's ratio (KR) in the entire sample group.

The findings of our study suggest that the SNPs rs404916326 T>A and rs408469734 G>A in the MSTN gene have the potential to serve as molecular markers for marker-assisted selection in sheep breeding, specifically for residual feed efficiency indicators.

Acknowledgements. The study was funded by the LZP-2021/1-0489 project: "Development of an innovative approach to identify biological determinants involved in the between-animal variation in feed efficiency in sheep farming."

Key words: sheep, feed efficiency, markers, DNA, myostatin, polymorphisms

Impact of serotonin concentration changes on the laterality of fruit flies (*Drosophila melanogaster* Meigen, 1830)

Tatjana Grigorjeva¹, Annija Kotova², Vita Viktorija Maļutina², Tatjana Krama^{1,3,4}, Sergejs Popovs^{1,4}, Māris Munkevics^{1,2,4}, Krists Zants², Ronalds Krams^{1,3,4}, Priit Jõers⁵, Giedrius Trakimas^{1,6}, Indriķis Krams^{1,2,4,7}

¹Daugavpils University, Institute of Life Sciences and Technologies, Daugavpils, LV-5401, Latvia; ²University of Latvia, Faculty of Biology, Rīga, LV-1010, Latvia; ³Estonian University of Life Sciences, Institute of Agricultural and Environmental Sciences, Tartu, 51006, Estonia; ⁴Latvian Biomedical Research and Study Centre, Rīga, LV-1067, Latvia; ⁵University of Tartu, Institute of Molecular and Cell Biology, Tartu, 51010, Estonia; ⁶Vilnius University, Institute of Biosciences, Vilnius, 10257, Lithuania; ⁷University of Tartu, Institute of Ecology and Earth Sciences, Tartu, 50409, Estonia. Corresponding authors e-mail: tatjana.grigorjeva@du.lv

The basic chemical reactions in the fruit flies (*Drosophila melanogaster* Meigen, 1830) brain are similar to those occurring in the human brain. In the flies' brain serotonin is responsible for the regulation of locomotion, sleep, general activity, light or shadow selection, as well as the regulation of intestinal tract activity and aggressiveness.

During the study we reared the wild type (Oregon-R) Drosophila flies, which were divided in 1 control group and 2 experimental groups, exposed to escitalopram and tryptophan added to their food. Escitalopram is an antidepressant and a specific serotonin reuptake inhibitor. This is a type of drug used to treat depression. In a long-term escitalopram reduces the release of serotonin in the brain. Tryptophan is an essential amino acid and a precursor of the neurotransmitter serotonin. Higher amounts of tryptophan in the body stimulate the increase of serotonin concentration.

The aim of the study was to analyse effect of escitalopram and tryptophan on the lateral movement variability in fruit flies. We used the Y-maze labyrinth method, which is commonly used to determine the effects of age, hormones, medicine, nutritional supplements and stress factors on the spatial short-term memory of the study object. During the trial, HD video camera with infrared sensor was placed above the Y-maze labyrinth plate and recorded all the fruit fly motions and turns made to the specific directions. The camera was connected with computer, which has the Noldus "EthoVision XT" video tracking software that registers all the turn coordinates.

The results showed that fruit fly lateral movement variability significantly increased in the flies from escitalopram exposure group. There was no significant difference in lateral movement variability between tryptophan exposure group and the control group flies. Y-maze is another effective method to study depressive behaviour of fruit flies and to understand the basic elements of depressive behaviour overall.

Key words: *Drosophila melanogaster*, Y-maze, serotonin, lateral movements, antidepressants, behaviour

Fauna of the predatory Mesostigmata mites (Acari, Parasitiformes) in the urban grasslands of Rīga city

Salmane, Ineta¹, Sniedze-Kretalova, Rūta²

¹Institute of Biology, University of Latvia, O. Vācieša iela 4, Rīga, Latvia;

²Latvian Fund for Nature,

Corresponding authors e-mail: ineta.salmane@lu.lv

Soil invertebrates, among them Mesostigmata mites, are known to be indicators of the state of the soil ecosystems including urban ones. Mesostigmata mites are dominant acarine predators playing a crucial role in soil food webs. An indicator of the degree of ecosystem degradation is the number of species and individuals and species composition. Soil Mesostigmata mites showing significant variability among natural and urban habitats. In collaboration with Latvian Fond for Nature and in the frames of the project "Introducing adaptive community based biodiversity management in urban areas for improved connectivity and ecosystem health urbanLIFEcircles" collection of soil samples in Rīga city grasslands was made. In total sampling was performed in 14 sites. Samples were taken by the soil corer and extraction of soil invertebrates on modified Berlese-Tullgren funnels was made. Although there was no intensive trampling in the investigated sites, grassland fragmentation un closeness of intensive traffic made a great impact on soil mite fauna. Mesostigmata mite species composition of investigated urban sites differed from that in the natural grasslands. Totally 20 Mesostigmata species in Rīga city grasslands were determined. Only six of those species were also known from the natural grassland habitats. The most frequent Mesostigmata species in urban grasslands were members of the families Rhodacaridae, Parasitidae and Laelaptidae. Mesostigmata species, known as eudominant in the territory of Latvia, were not found in the urban grassland soils of Rīga city.

Key words: predatory Acari, urban habitats, species composition

References

Salmane, I., Brūmelis, G. 2010. Species list and habitat preference of Mesostigmata mites (Acari, Parasitiformes) in Latvia. Acarologia 50/3: 373-394.

Grina, V., Kagainis, U., Juceviča, E., Salmane, I., Melecis, V. 2023. Soil microarthropod distribution on the urban–rural gradient of Riga city: a study with robust sampling method application. Journal of Urban Ecology 9/1, juad012, https://doi.org/10.1093/jue/juad012

Koehler, H. 1999. Preadatory mites (Gamasina, Mesostigmata). Agriculture. Ecosystems and Environment 74: 395–410.

https://grasslife.lv/darbi/plavu-atjaunosana/pilsetas-plavas

Abstracts Poster Presentations

Microsatellite Genotyping of *Cyprinus carpio* Czech and Lithuanian Strains

Gabriela Liubartaite¹, Dr. (HP) Dalius Butkauskas¹

Nature Research Centre, Akademijos St. 2, LT-08412, Vilnius Corresponding authors e-mail: gabriela.liubartaite@gmail.com

Several years ago, our laboratory has begun research looking into the genetic diversity of Lithuanian common carp strains with the aim of potentially providing aid with maintaining a healthy carp gene pool in Lithuania. Common carp samples were collected from 5 different Lithuanian farmlands, alongside an additional Czech strain carp population to serve as a comparative control, totaling to 142 samples and 5 distinct carp populations. For the assessment of genetic diversity within each population, a set of 8 microsatellite loci was selected. Across all 5 populations, a total of 85 alleles were determined, with the highest mean allele count per population being 7.3, and the lowest – 3.3. Lowest expected heterozygosity across all populations was found to be 0.57, the highest - 0.62, while lowest and highest observed heterozygosities have been found to be 0.60 and 0.71, respectively. Furthermore, molecular variance analysis (AMOVA) revealed that the majority of molecular variance - 76% - is found within populations, with the remainder of variance being between populations, which may be due to the structural differentiation between the Lithuanian and Czech strains. This strain-specific clustering of variance was further observed after conducting a principal coordinate analysis (PCoA), wherein Lithuanian-strain samples were found to cluster between each other, suggesting a high level of gene flow between the farms, while the Czech-strain individuals formed a genetically distinct cluster, thus showcasing a distinct, strain-specific genetic structure, and indicating a possibility to discriminate carps of Czech origin and Lithuanian origin based on genotyping using microsatellite markers.

Key words: *Cyprinus carpio*, carp strains, genetic diversity.

Molecular Investigation of *Sarcocystis* spp. in Invasive Raccoon (*Procyon lotor*) from Lithuania

Evelina Maziliauskaitė, Dalius Butkauskas, Evelina Juozaitytė-Ngugu, Živilė Strazdaitė-Žielienė, Petras Prakas.

Nature Research Centre, Vilnius, Lithuania

Corresponding authors e-mail: evelina.maziliauskaite@gamtc.lt

The raccoon (*Procyon lotor*) is a relatively small carnivorous mammal belonging to the family *Procyonidae*. In native areas, raccoons are known to be hosts of many parasites that can be transmitted to humans, domestic and wild animals. The apicomplexans of the genus *Sarcocystis* are hazardous cosmopolitan parasites spread by meat consuming carnivores, omnivores, and scavengers, such as raccoons. The raccoon was introduced from North America to Europe in the 20th century, and since 2010 it has been spreading in Lithuania. The increased raccoon populations and their geographical spread may lead to new disease threats in Europe and Lithuania.

From 2020 through 2023, 13 raccoon samples were collected from north-western Lithuania. In this work, the raccoon was studied as a potential definitive host of *Sarcocystis* parasites by examining the small intestines of these animals. Sedimentation of intestinal mucosal scrapings was performed to search for *Sarcocystis* spp. oocysts/sporocysts. The morphology of sporocysts and oocysts of distinct *Sarcocystis* species does not differ, these cysts overlap and co-infections of *Sarcocystis* spp. frequently occurs in a definitive host. Therefore, *Sarcocystis* species were identified by molecular methods.

By light microscopy, free sporocysts and sporulating oocysts of *Sarcocystis* spp. were noticed in seven (53.85%) of 13 animals tested. Based on the amplification of 28S rRNA, ITS1, and cox1 regions and subsequent sequencing, Sarcocystis spp. were detected in five samples. Notably, more than two *Sarcocystis* species were established in four animals. Sarcocystis spp. using birds or predatory mammals as their intermediate hosts and birds as definitive hosts were identified in three raccoons. Two samples contained Sarcocystis spp. circulating between rodents and birds. *Sarcocystis rileyi* producing macroscopic sarcocysts in the muscles of ducks were detected in the intestines of four animals. Finally, *S. bovifelis* infecting cattle and two yet undescribed *Sarcocystis* sp. closely related to parasites forming sarcocysts in muscles of ungulates were identified in two raccoons. The results show that invasive raccoons are important omnivores for transmitting various *Sarcocystis* spp.

Key words: Sarcocystis, molecular identification, raccoon

Intraspecific genetic variability of Cloudberry (*Rubus chamaemorus*) in the Eastern Baltic region based on microsatellite data analysis

Viktorija Levinger¹, Dace Grauda², Nikole Krasnevska², Andra Mikelsone², Olesja Kruchonok, Dalius Butkauskas^{1,2}

Corresponding authors e-mail: viktorijalevinger20@gmail.com

Cloudberry (Rubus chamaemorus) is a perennial plant native to the northern hemisphere. Its berries are rich in vitamin C and antioxidants and therefore have a high nutritional value. It is used in traditional medicine to treat scurvy and diarrhea and is grown on an industrial scale in Scandinavian countries. In this work, 111 different clones representing wild population of R. chamaemorus were studied. The clones were collected from 18 wetlands: 2 from Lithuania, 12 from Latvia and 4 from Belarus. The aim of the study was to determine the extent of inter-population differentiation and intrapopulation genetic diversity of R. chamaemorus distributed in the continental zone of the Eastern Baltic region and northern part of Belarus. Initially 8 primer pairs specific for blackberry (Rubus caesius), raspberry (Rubus arcticus) and cloudberry (R. chamaemorus) were screened for suitability to be applied in high-throughput microsatellite analysis. Six primer pairs enabled amplification of polymorphic fragments and were further used on study genetic variation within and among populations. From 2 to 6 alleles per locus and a total of 104 different genotypes were identified. Two genetically differentiated subpopulations were distinguished based on structure analysis. These sub-populations are distributed in the Western and Eastern parts of the studied area being separated from each other by the Central Lithuanian Lowland and the Zemgale Plain. Some of cloudberry plants appeared more genetically similar to the majority of individuals attributed to other subpopulation and it could be associated with possible transfer of seeds from one wetland to another by migrating animals.

Key words: cloudberry, microsatellites, genetic analysis, subpopulations.

¹ Nature Research Centre, Vilnius, Lithuania

²University of Latvija, Institute of Biology, Riga, Latvija

Optimization of oat (Avena sativa L.) embryo culture method for breeding

Krasnika Nadežda, Miķelsone Andra, Grauda Dace Institute of Biology, University of Latvia Corresponding authors e-mail: andra.mikelsone@lu.lv

Oat (Avena sativa L.) embryo culture can be widely used in modern breeding, to speed up the breeding processes. Seedlings can be obtained at different stages of seed development: from premature to mature and also from practically non-germinating seeds. The composition of the medium, including the concentration of sucrose, can significantly affect the growth and development of oat embryos, thereby increasing the percentage of viable plants. The aim of this study was to determine the optimal medium composition for oat embryo cultures in order to accelerate their growth and development.

Two types of medium, B5 (Gamborg's B5 Medium) and MS (Murashige and Skoog) with three different sucrose concentration: 1%, 2% and 3%, were used. Oat seeds were sterilized and then the embryo was removed. All embryos were placed in tubes with medium and incubated in the grow chamber (24 °C, photoperiod 16/8 h) and the leaf and root size of each plant was measured every day. After nine days plantlets were transferred to soil. After 14 days of cultivation the length of leaves and roots was measured, as well the number of leaves and roots was counted for each plant. It was observed that the most suitable medium for oat embryo culture was MS with 3% sucrose; after one day of cultivation on this medium, root germination was observed in 64% of all embryos and leaf germination – 52%. Also, after nine days of embryos cultivation on MS medium with 3% sucrose, the best result was observed, 57% of plants were suitable for planting in soil.

Key words: embryos culture, oat, breeding

Preliminary results of soil mesofauna characterized by QBS index in the background of restoration success of urban grasslands in Riga city, Latvia

Ineta Salmane¹, Edīte Juceviča¹, Uģis Kagainis¹, Rūta Sniedze-Kretalova ² ¹Institute of Biology, University of Latvia, O. Vācieša iela 4, Rīga, Latvija ²Latvian Fund for Nature, ruta.sniedze@ldf.lv Corresponding authors e-mail: edite.jucevica@lu.lv

Soil arthropods are considered as one of the most effective bioindicators to assess soil health and biological properties that are important in plant development and sustainability. A grassland management program has recently been launched in order to restore natural meadows in the city of Riga where soil arthropod communities are planned to be used as a co-characteristic unit for further assessments during the program. In correspondence to the alternative methodology by Italian authors (Parisi et al., 2005) soil arthropods were used as soil management bioindicators without identifying their species in a considerably faster and easier manner. By classifying their biodiversity through their ecomorphological index (EMI) typology a sum of points (QBS - "Qualità Biologica del Suolo", i.e. Biological Quality of Soil) we characterised the investigated plots of grasslands.

Eleven urban meadows were selected in the city of Riga and grouped according to their degree of degradation. Soil arthropods were collected at the end of October 2023. In each sample plot ten soil cores (5 cm diameter, 15 cm depth) were taken along the 30 m transect. Soil animals were extracted from soil samples using a Berlese-Tullgren extractor. EMI and QBS indexes were determined for each sample. EMI scores range from 1 to 20 for each group, and QBS is the sum of EMI points in the sample.

QBS values correlated with environmental conditions among different urban meadows by Spearman and Kendall tests. In PCA analysis, the most severely degraded plots among those of the central part of the city with intensive traffic roads nearby significantly corresponded to lower QBS. Factors e.g. relative soil moisture, vegetation height and naked soil cover described the variation of QBS and plant species for the most part of variation data for the first two axes (20.6-55.9%). We believe that further surveys at the sampling sites will improve the analysis, and the QBS evaluation method is expected to give a more precise assessment of how successful the urban meadow restoration program was.

Key words: Urban grassland, Soil arthropods, QBS

Parisi V., Menta C., Gardi C. Jacomini, C. Mozzanica, E. 2005. Microarthropod communities as a tool to assess soil quality and biodiversity: a new approach in Italy. Agriculture Ecosystem and Environment 105, 323-333.

Effect of treatments with microalgae extracts to total chlorophyll content in tomato leaves

Authors: Irina, Sivicka¹; Kaspars, Kampuss¹; Pāvels, Semjonovs²; Ingrīda, Augšpole¹. Affiliation: ¹Institute of Soil and Plant Sciences, Latvia University of Life Sciences and Technologies; ²Laboratory of Industrial Microbiology and Food Biotechnology, Institute of Biology, University of Latvia

Corresponding authors e-mail: Irina.Sivicka@lbtu.lv

The research aimed to the evaluation of total chlorophyll content in tomato leaves under the treatment with microalgae extracts. In August 2023, tomato seedlings (cultivar 'Belle' F1, Enza Zaden) were planted in 25 L pots, filled with peat (producer Laflora LTd., pHKCl 5.5) and grown in polycarbonate greenhouse of the laboratory of Horticulture and Beekeeping of the Latvia University of Life Sciences and Technologies. The plants were sprayed weekly till with the solution of ethanol extractions of different microalgae species: Spirulina, Dunaliella, Chlorella, from 14th August till 11th September, till harvesting time, total 5. In total, 9 plants per treatment were used. Two concentrations of the extracts were compared with sprays with corresponding ethanol solution as a control. With a low-cost handheld chlorophyll meter atLEAF+, after the treatments, at Leaf index (describing plant health, based on chlorophyll contents in leaves), was measured. The measurements were made on each level of plant (top, middle and lower internode). The total chlorophyll content, mg cm-2, in tomato leaves was obtained by converting the atLEAF CHL values in SPAD and considering the relationship among chlorophyll content and SPAD units. As well as correlation with tomato leaves' length and width was counted.

During experiment, for any microalgae extracts no negative effects to tomato plants' growth and development were noticed. The total chlorophyll content varied from 0. 287 mg dm-2 (control variant with drinking water) till 0.490 mg dm-2 (treatment with Dunaliella, 10% concentration). Average result for all variants was 0.383 mg dm-2.

It was observed, that influence of microalgae extract`s type as well as concentration to the total chlorophyll content was not significant (p > 0.05). In the case of level of plant, where measurements were made, the significant influence (p < 0.05) on the total chlorophyll content was observed. No correlation with tomato leaves` parameters was found.

Acknowledgments: This study was performed within the framework of the project No. 22-00-A01612-000014 co-financed by European Agricultural Fund for Rural Development (EAFRD) and supported by the Ministry of Agriculture and Rural Support Service of the Republic of Latvia.

Key words: chlorophyll, tomato, microalgae, extracts

Mixotrophic cultivation of microalgae in whey medium for poultry diet supplementation with microalgal biomass

Authors: Kristaps Neiberts, Pāvels Semjonovs

Laboratory of Industrial Microbiology and Food Biotechnology, Institute of Biology,

University of Latvia

Corresponding authors e-mail: kristaps.neiberts@lu.lv

Just as global human population is increasing every day, the demand for variety of different goods also increases. To be able to deliver enough goods for human society, we need to expand research not only on land, but also in freshwater and saltwater bodies. There is an aquatic group of microorganisms that is drawing favourable attention of scientists – microalgae.

Poultry farming has a great impact on delivering human diet with eggs and meat. To have high quality eggs and meat, high-quality diet should be provided for poultry, which is rich in lipids, polyunsaturated fatty acids, proteins, vitamins, complex carbohydrates, and other functional compounds.

Microalgae are microorganisms that can be used to fulfil those requirements. It has been shown that, to increase microalgae biomass yield, it is possible to cultivate microalgae mixotrophically in whey medium. Besides, by cultivation in whey medium there can be seen decrease in ecological impact of improper disposals, as whey considerably composes most important part of dairy processing by-products. But not all microalgae are able to consume lactose, which is the main sugar in whey. So there needs to be found microalgal strains which are able to consume lactose as additional carbon source during mixotrophic cultivation.

In this research have been obtained environmental isolates and microalgal strains have been screened for lactose assimilation properties. Isolated strains have been identified as *Graesiella emersonii* and Tetradesmus obliquus as well as unidentified isolate X1, and compared with *Chlorella vulgaris* CCAP 211/11 as commercially already widely used microalgae and other cultures - Scenedesmus quadricauda CCAP 276/16, *Chromochloris zofingiensis* CCAP 211/14, *Galdieria sulphuraria* UTEX 2919. Heterotrophic growth pattern of *G. emersonii* and *T. obliquus* showed highest biomass yield compared to other evaluated microalgal cultures when grown in presence of lactose, however *C. zofiengiensis* showed greater biomass yield as compared to G. sulphuraria, which could be related to higher lacZ gene activity. By performing further mixotrophic growth experiments in whey, both environmental isolates and C. zofiengiensis showed higher biomass productivity then in photoautotrophic control. Results confirm that during mixotrophic growth in whey it is possible to obtain a higher biomass yield then during photoautotrophic cultivation.

In further experiments optimisation of whey media should pe performed to obtain greater impact of mixotrophic cultivation on selected microalgal strains biomass productivity and yield. As well as it should be evaluated further whether changes in media composition can also provoke the changes in microalgal biomass composition.

Key words: microalgae, Graesiella emersonii, Tetradesmus obliquus, whey

This study was performed within the project "Development of plant origin feed supplement for strengthening poultry immunity and increasing nutritional value of eggs with omega-3 fatty acids (grant Nr.: 22-00-A01612-000015) co-financed by European Agricultural Fund for Rural Development (EAFRD) and supported by the Ministry of Agriculture and Rural Support Service of the Republic of Latvia.

Optimization of the production of sodium alginate derivatives

Svetlana Vasiljeva¹, Roberts Krumins², Galina Smirnova^{1,3}, Natalija Basova¹, Abbas Mirshafiey⁴, Dmitry Babarykin²

¹University of Latvia, Institute of Biology, Riga, Latvia; ²Faculty of Biology, University of Latvia, Riga, Latvia; ³Institute of Innovative Biomedical Technology Ltd., Riga, Latvia; ⁴ Italian Multiple Sclerosis Society Foundation (FISM), Genoa, Italy

Correspondence: svetlana.vasiljeva@lu.lv

Sodium alginate, a salt of alginic acid, is a linear polysaccharide consisting of two forms of linked hexuronic acid residues - d-mannuronic acid (MA) and I-guluronic acid (GA). The polysaccharide found predominantly in brown algae and widely used as a wetting and thickening agent in the food, pharmaceutical, cosmetics and other fields. Until recently, MA and GA were used in scientific research, including the assessment of their effectiveness as a plant growth stimulator. A significant impetus to the study of the biological effects of MA and GA was given by the discovery of their ability to act as a non-steroidal anti-inflammatory agent with immunomodulatory effects.

Thanks to A.Mirshafiey scientific study, a created method for MA and GA synthesizing from sodium alginate was developed and patented (PCT/EP2023/087660, PCT/EP2023/087659, EP/22.216780.1). The method is based on the hydrolysis of sodium alginate with sulfuric acid followed by the separation of MA and GA, that included Ph control, centrifugation and quite long-time drying process. The method is rather complex, time-consuming and energy-intensive. The activity of these two uronic acids was assessed separately. Having compared the biological effects of MA and GA, only non-essential differences between them were established. At the same time, MA was the main necessary product, while GA almost was not used. This suggested the possibility of practical use of a mixture of MA and GA without prior separation.

The purpose of this study was to optimize the previously created method for the production of uronic acids aimed at improving the cost effectiveness of the technology.

The MA+GA mixture obtained as a result of technological process parameters correction does not require pre-drying before subsequent use. It has higher stability compared to dry MA and Improved the consumer properties. The modified technology increases product yield by 12.0±3.0 % (65.6±4.3 vs 53.8 ±3.1%), reduces the technological process duration by 6 h, the number of production steps, drying time, the consumption of components and the formation of waste requiring special disposal. At the same time, the CE indicator for the production of the uronic acid composition reduced by 36%. The effectiveness of finished product made on the basis of the MA+GA mixture was the same as in the case of MA. The optimization of the technology for the synthesis of sodium alginate derivatives makes it possible to increase the economic efficiency of the process. The composition of uronic acids obtained in this way can be used in the creation of new innovative products for human health.

Key words: mannuronic acid, guluronic acid, polysaccharide composition

P9

The anti-inflammatory effect of Mannuronic acid (M2000) in experimental model of glomerulosclerosis

A. Mirshafiey ^{1,2}, A. Sahmani, P. Ekhtiari, Z. Aghazadeh

¹ Int.Reviewer/FISM (Italian Multiple Sclerosis Society Foundation); ²Livonian Biotech Millennium (LBM), Riga, Latvia

Corresponding authors e-mail: mirshafiey@tums.ac.ir

The anti-inflammatory effect of a patented (DE-102016113018) novel natural agent, Mannuronic acid (M2000) extracted from Sodium Alginate was tested in experimental glomerulosclerosis, as a model of nephropathy.

The Adriamycin was used for induction of experimental glomerulosclerosis, the animal model of nephrotic syndrome. To induce glomerulosclerosis, Adriamycin was given once by a single intravenous injection (7.5 mg/kg) through the tail vein. Six days after injection of Adriamycin, the intraperitoneally (i.p) injection of 30 mg/kg Mannuronic acid solution was administered. The total of i.p. injections were 14, in which five injections were carried out every day and nine injections were done at regular 48-h intervals. This experimental protocol was terminated on day 28 and animals were killed six weeks after the induction of disease. The treated patient rats revealed a significant reduction in serum creatinine, proteinuria, BUN, and serum cholesterol. Moreover, the i.p. injection of Mannuronic acid could significantly reduce the serum level of interleukin-6 in treated animals in comparison to non-treated controls. In addition, the i.p. administration of Mannuronic acid significantly reduced number of glomerular leukocytes, hypercellularity and hydropic change in capillary network within the renal cortex and diminished tubular casts.

Conclusion: These findings showed that treatment with Mannuronic acid (M2000), as a novel natural anti-inflammatory agent can reduce proteinuria, and suppress the progression of glomerular lesions in animal model of glomerulosclerosis.

Key words: Mannuronic acid; animal model of glomerulosclerosis, proteinuria

Anti-tumoral effects of Guluronic acid (G2013), as a novel safe natural agent on animal model of breast cancer

A. Mahdian-Shakib, H. Hassannia, F. Hosseini, F. Jadidi-Niaragh, P. Kokhaei, Abbas Mirshafiey^{1,2}

¹ Int.Reviewer/FISM (Italian Multiple Sclerosis Society Foundation); ² Livonian Biotech Millennium (LBM), Riga, Latvia

Corresponding authors e-mail: mirshafiey@tums.ac.ir

A continuous chemical, physical and mechanical irritation and following chronic inflammation could be led to a tumoral tissue as well as malignant progression in several cancer types. This investigation was conducted to evaluate the potency of a very safe natural anti-inflammatory agent, Guluronic acid (G2013) on experimental model of breast cancer. Our findings showed that Guluronic acid is able to inhibit significantly the cancer related inflammation as well as tumor-promoting mediators (MMP2, MMP9, VEGF, COX-2 and also pro-inflammatory cytokines) without cytotoxic effects. In addition, Guluronic acid (G2013) could significantly inhibit the tumor cell adhesion to extracellular matrix and reduce accumulation of inflammatory cells in tumor-bearing mice. The results of this investigation were associated with decreased tumor growth, metastasis, angiogenesis and prolonged mice survival.

Conclusion; the present research showed that the Guluronic acid (G2013) as a novel natural anti-inflammatory agent is able to inhibit tumor growth, metastasis and angiogenesis and acts as anti-tumoral agent which might be considered as a novel promising strategy for cancer prevention and treatment.

Key words: natural anti-inflammatory agent; tumor-promoting mediators; cancer prevention

Inhibitory property of Mannuronic acid on angiogenesis phenomenon using experimental model

Mohsen Rastegari-Pouyani, Ali Mostafaie, Abbas Mirshafiey^{1,2} Int.Reviewer/FISM (Italian Multiple Sclerosis Society Foundation); ² Livonian Biotech Millennium (LBM), Riga, Latvia Corresponding authors e-mail: mirshafiey@tums.ac.ir

Angiogenesis or neovascularization is a biological phenomenon which contributes significantly in pathogenesis of inflammatory diseases and solid tumors growth, so that any agent with anti-angiogenesis property is able to reduce the inflammatory reactions and inhibit tumor growth process. The Mannuronic acid (M2000) is a new very safe natural anti-inflammatory agent and a novel metalloproteinases (MMPs) inhibitor. This investigation was conducted to determine the anti-angiogenesis property of Mannuronic acid under examinations of in vitro and in vivo. These experiments were carried out based on the 3D collagen-cytodex model and the chick chorioallantoic membrane (CAM) assay for evaluating the anti-angiogenesis effect of Mannuronic acid (M2000). Cytotoxicity examination showed that Mannuronic acid, at concentrations of less than 100 µg/mL had no cytotoxic effect on human umbilical vein endothelial cells (HUVECs). It was also revealed that, the anti-angiogenesis effects of M2000 is marginal at in vitro model as well as significant and dose-dependent at in vivo status. This research indicated that M2000 might be recommended as a novel natural antiangiogenic molecule which essentially exerts its activity mainly via indirect effects on endothelial cells and its anti-inflammatory property may partly be associated with its anti-angiogenic activity. Thus, Mannuronic acid (M2000) might be considered as a novel natural agent for prevention and treatment of cancer, inflammatory diseases, and other angiogenesis-related disorders.

Key words: neovascularization; cytotoxicity; metalloproteinases (MMPs) inhibitor

Different model systems to evaluate EMR impact

Kārlis Žagata, Elīna Ažēna, Dace Grauda University of Latvia, Institute of Biology, Jelgavas str. 1, Rīga, Latvia Corresponding authors e-mail: karlis.zagata@lu.lv elina.azena@lu.lv

Electromagnetic radiation (EMR) pollution caused by such sources as UV-B rays or electromagnetic field (EMF) generated by electric equipment or both combining has become as one of the most important aspects of urban environmental in nowadays. Therefore, using materials endowed with properties to help protect against EM radiation is one of best way can be minimized EMR influence. But how to determine effectiveness of protective properties of these new materials is still actual – model systems for short- and long-term impact of EMF should be sensitive, give reliable and quickly obtainable results.

In this work as the model system to evaluate short-term impact of EMR were choose two model organisms: 1) yeast (*Saccharomyces cerevisiae*) liquid culture and 2) fruit fly (*Drosophila melanogaster*) larvae and two different types of EMR sources: a) UV radiation with ray emission at 290-315 nm (UV-B) and radiation intensity 5.000 mW/m2 and b) experimentally generated EMF environment using Helmholtz coil by alternating current frequency 50Hz and field density 500 µT.

It is known that formation of reactive oxygen species (ROS) can be stimulated by a variety of stressors incl. electromagnetic radiation, and this phenomenon was used in assessment of efficiency of chosen model systems. 2',7'-Dichlorodihydrofluorescein diacetate (C24H16Cl2O7) as marker of EMR influence was used – intracellularly deesterified it turns to highly fluorescent 2',7'-dichlorofluorescein upon oxidation. Fluorescence was excited at wavelength 385 nm and detected at 435 nm by Tecan Spark® multimode reader.

By using of UV-B rays as the stress factor on yeast liquid culture effect was detected already after 30 minutes, maximum impact was observed after 60 minutes. Fluorescence of yeast culture impacted by UV-B rays was dramatically (470%) higher comparing to control. By using of EMF of density 500 μ T as the stress factor no influence of EMF on yeast culture was observed. A similar reaction to UV-B was observed in a fruit fly model where the fluorescence of larval homogenates increased by 176% after 75 minutes of irradiation.

In conclusion both model organisms (Saccharomyces cerevisiae and Drosophila melanogaster) are usable to detect the protective properties of materials against UV-B rays but usage for investigations of influence of EMF in density of 500 μ T unfortunately is questionable.

Acknowledgments: The research is supported by project Nr. ES RTD/2022/7 "3D Biotextile with Technological Composition of nano particles to enhance the protecting properties" (Latvia).

Key words: Saccharomyces cerevisiae, Drosophila melanogaster, UV-B, EMR, ROS