symbioza

Intercollegiate
Biotechnology
Symposium

The 10th Prof. Krzysztof W. Szewczyk

Intercollegiate Biotechnology Symposium

SYMBIOZA

BOOK of ABSTRACTS

12–14 May 2023, Warsaw & online

The 10th Krzysztof W. Szewczyk
Intercollegiate Biotechnology Symposium "Symbioza"
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Warsaw, 12 May 2023

Dear Participants,

I am delighted to welcome you to the 10th edition of Intercollegiate Biotechnology Symposium "Symbioza". After a long break, we are finally able to meet face-to-face to share our views on the fascinating world of science.

It is worth noting that evolution is not limited to living organisms. The Warsaw Society of Biotechnology "Symbioza"has undergone significant changes over the past decade since its inception. Initially, the Society's main focus was organizing an annual Symposium. However, its members have since expanded their horizons, engaging in various activities to bring together students, the biotech industry, and the scientific community in Warsaw. Even the Society's oldest idea, the Intercollegiate Biotechnology Symposium, has not remained static. In 2021, due to the global pandemic, the conference took on a radically different form, transitioning to an online event.

With the help of members who have participated in organizing previous editions of the conference, the 10th anniversary edition of the Symposium successfully combines traditional and new methods of event organization. Hosting the event in a hybrid format enables biotechnology enthusiasts who are unable to attend in-person to participate and benefit from the rich scientific content. We have also decided to use the livestream to demonstrate our support for young biotechnologists from Ukraine by allowing them to participate online, free of charge. We hope to have the opportunity to gather in person once again soon.

The pandemic restrictions have taught us to value in-person meetings even more. With this in mind, we strive to provide Symposium participants with various experiences, not only through lectures covering a broad range of biotechnology topics, but also through social activities, including an outdoor game. Our aim for "Symbioza" is to offer many memorable moments.

While we hope you enjoy the social aspects of the Symposium, we remain committed to the primary goal of the event: to share knowledge and serve as a bridge between students, scientists, and the industry. Aspiring young biotechnologists face numerous career choices, and with so many pressing world problems and research areas to explore, any guidance can be valuable. We cordially invite you to attend the first-ever discussion panel in the history of "Symbioza" where our esteemed guests will dispel doubts about different career paths in biotechnology.

Furthermore, participants will have the opportunity to attend lectures given by six remarkable keynote speakers, four representatives of biotech companies, and thirty young scientists. The presentations will be divided into eight oral sessions with particularly charming names. On-site attendees will also have the chance to explore additional fascinating research topics and view participants' posters during two poster sessions.

10th IBS "Symbioza" is brought to life by remarkable individuals including lecturers, young scientists sharing their research insights, a curious audience, and the numerous members of the Warsaw Society of Biotechnology "Symbioza" who worked tirelessly day and night to make this event possible. I would like to conclude this welcome message by expressing my deepest gratitude to each and every member of the Organizing Committee who dedicated their time and efforts to organize the 10th Intercollegiate Biotechnology Symposium "Symbioza".

— **Olga Kowalska**

President of the Warsaw Society of Biotechnology 'Symbioza'

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It is pointless to indicate which parts of technology or activities are more important. What is necessary, however, is mutual understanding of cooperating specialists.

— Krzysztof W. Szewczyk

Prof. Krzysztof Włodzimierz Szewczyk (1952–2011) was a remarkable scientist, and a well-recognized specialist in the fields of industrial biotechnology and bioprocess engineering. He co-founded and organized biotechnology studies at Warsaw University of Technology (WUT). He was also a director of the Intercollegiate Biotechnology Centre at WUT (2007–2008) and a supervisor of the Department of Biotechnology and Bioprocess Engineering at the Faculty of Chemical and Process Engineering at WUT (2006). Since 2003 he had been a member of Committee of Biotechnology during the Presidium of Polish Academy of Sciences, the secretary of the Bioprocess Engineering section in the Committee of Chemical Process Engineering at Polish Academy of Sciences (1992–1995), a member of Programme Council of the "Biotechnology" quarter journal (2005–2010), and a Vice-President of Polish Federation of Biotechnology (2007–2010).

Prof. Szewczyk was an author of more than 120 scientific articles, co-author of 6 patents and utility designs and a co-author of 8 student handbooks. He was known as an excellent and valued teacher among students not only at his alma mater, but also at the University of Warsaw, where he taught bioprocess engineering. In 1995, he received the Silver Cross of Merit, and in 2003 he was awarded with the Commission of Education Medal and in 2008 distinguished with Ministry of Science and Higher Education Award. His colleagues, fellow professors and students remember him as an erudite, a classical music lover, and a chess enthusiast who was truly wedded to education among academic adolescents.

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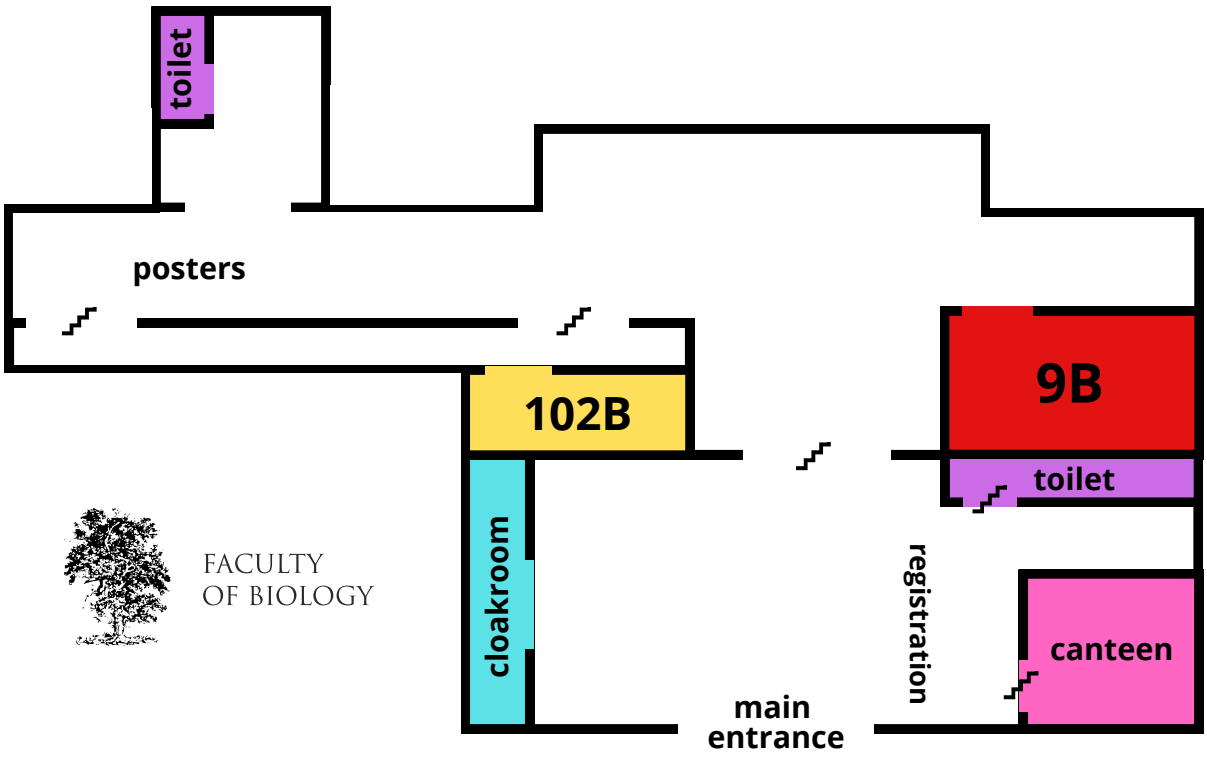
Conference Venue

Faculty of Biology of the University of Warsaw – the best biology programme in Poland. The University of Warsaw was ranked first in all study programmes available at Faculty of Biology in *Perspektywy* ranking 2021.

The Faculty of Biology of the University of Warsaw includes eight institutes, a botanic garden, three field stations, and five laboratories/core facilities. It hires nearly 400 employees, including ca. 200 academic staff. Each year, about 500 students learn biology, biotechnology and environmental protection, while ca. 120 graduate students develop their PhD projects.

We are more than pleased to boast about the honorary patronate of Faculty of Biology since this year's edition takes place in Faculty's building!



Floor plan

Organizer

Warsaw Society of Biotechnology 'Symbioza' (WSB 'Symbioza') was established in 2013, thanks to cooperation between students from three major Warsaw universities: University of Warsaw, Warsaw University of Life Sciences and Warsaw University of Technology. WSB 'Symbioza' brings together young life sciences enthusiasts with a particular interest in biotechnology who want to share knowledge and experience with students and scientists coming from around the world. The main purpose of the Society is popularisation and increase of consciousness in society about biotechnology by organizing a wide range of activities.

The Intercollegiate Biotechnology Symposium 'Symbioza' (IBS 'Symbioza'), being the Society's oldest and most important event, is a scientific conference addressed to international students and PhD candidates who want to broaden their knowledge on biosciences. For many, this is the first opportunity in their research careers to present their findings to wider audience. The symposium serves also as a forum to network with young scientists around the world, which leads to successful collaborations, internships, etc. The 8^{th} edition of IBS 'Symbioza' was awarded in the StRuNa Competition as 'Conference of the year 2019'.

Feel the Flow is a project which aims to popularize science among the public in an informal and friendly atmosphere, as well as discuss issues that people face pursuing scientific career. This is realized with a series of meetings (recently online) with renowned science communicators from different branches of life sciences – from physics and chemistry through biology to medicine.
BARWy (Biotechnological Aspects of Various Choices) is an event aimed to show high school graduates and freshmen biotechnology students a variety of professional perspectives and possibilities awaiting them in their future careers, be it in industry or academia. During the event, there is a remarkable opportunity to meet representatives of different biotechnology related professions and listen to their successful stories.

During *OAKs (Attractive Conventicles Camps)*, several days long retreats, attendees take part in workshops that aim to improve the scientific presentation and communication techniques as well as show new ways of transferring knowledge and presenting research results.
WSB 'Symbioza' is also actively participating in the yearly *Science Picnic* of Polish Radio and the Copernicus Science Centre in Warsaw. During a family friendly whole day event with thousands of visitors, we strive to uncover and explain the fascinating world of biotechnology to the youngest enthusiasts.

Symbioza Umysłów (*Symbiosis of Minds*) is the latest project of the Society that is addressed to secondary schools students. During the recent COVID-19 pandemic, when most education was held online and remotely, we organized an interdisciplinary competition aiming to popularise science and promote cooperation and synergic thinking across the borders among people and scientific disciplines.

Organizing Committee

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Aleksander Lempert — Treasurer, Head of Logistic Section
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Co-organizers

The organizers of the 10th IBS 'Symbioza' have been supported by four biotechnology student organizations affiliated at the four most important Warsaw universities.

"Antidotum" is a student organization that has been active at the University of Warsaw since 2012. Over the years, the organization has become an important platform for students to explore and deepen their knowledge in the field, as well as exchange experiences with like-minded individuals. One of the most notable achievements of the organization is the successful organization of the "Biofusion" student scientific conference. This conference has been organized for five editions so far, and has attracted significant interest from students and academics alike. Through its activities, "Antidotum" has become a hub for students interested in biomedical sciences at the University of Warsaw. Overall, "Antidotum" remains a vital and important student organization at the University of Warsaw, providing a supportive community and a platform for students to engage in meaningful discussions, exchange ideas, and explore the exciting and ever-evolving field of biomedical sciences.

Biotechnology Student Association "Herbion" was established in 2003 at the Faculty of Chemistry of Warsaw University of Technology. "Herbion" carries out a number of scientific projects, which currently include the cultivation of lactic acid bacteria on coffee waste in an air-lift bioreactor, along with studying the influence of various substrates on beer production. We also popularize biotechnology through science shows and mass events such as the Science Picnic of Polish Radio and the Copernicus Science Centre, open days at Warsaw University of Technology, and others. In addition, we organize science workshops for aspiring scientists from primary and middle schools. To help students jumpstart their scientific careers, we also take part in organizing the annual life science job fair "SSP"and help with finding projects for scientific volunteering. Other activities include educational trips and organizing monthly online lectures, known as *"Meetoza – dzielimy się wiedzą"*, some of which are available in English on our Facebook page.

KNBiotech Science Club is a student research organization at the Faculty of Biology and Biotechnology at the Warsaw University of Life Sciences (SGGW). Operating since 1997,the club unites students of biological faculties interested in the broadly understood biological sciences, in particular focusing on the field of biotechnology. Members carry out specific projects aimed at developing their interests. In addition to scientific activity, participants of KNBiotech are involved in popularization events, such as the Science Picnic of Polish Radio and the Copernicus Science Center, Days of Warsaw University of Life Sciences and numerous scientific conferences.

The Students Research Club EVIONA is a community of young scientists who are interested in the biology of Extracellular Vesicles (EVs) and their functions as mediators in various diseases. Their community was established in September 2022, and they work under the supervision of Małgorzata Czystowska-Kuźmicz in the Department of Biochemistry at the Medical University of Warsaw. EVIONA's aim is to encourage young and prominent scientists to study EVs and provide them with the opportunity to work in the research field from the first years of their studies. We are particularly interested in the role of EVs in immunology, oncology, and metabolic diseases. The field of EVs is constantly expanding, and we believe that everyone can find something of interest in this field!

The Warsaw Society of Biotechnology "Symbioza" proudly stands in solidarity with our Ukrainian counterparts in light of the human rights violations and unjustified Russian aggression they have faced. The Organizing Committee of the Symposium is committed to actively supporting the researchers and students from Ukraine, and we are pleased to announce a special initiative to demonstrate our solidarity.

In recognition of the challenges faced by the Ukrainian society, including researchers, we declare that remote participants with Ukrainian affiliations are exempt from the conference fee. This exemption serves as a symbolic gesture, demonstrating our support and ensuring that our Ukrainian colleagues have equal opportunities to participate and contribute to the symposium. As we gather at the symposium to celebrate advancements and innovation in biotechnology, we remain mindful of the challenges faced by our colleagues in Ukraine. Through this small gesture and ongoing efforts, we strive to strengthen the bond between our two nations, promoting academic and cultural exchange while standing together in the face of adversity.

We believe that together we can build a brighter future founded on cooperation and symbiosis.

Program overview

Day 1: Friday	from 13.00	Registration	
	15.00–15.15	Opening of the Conference	
	15.15–16.05	■ **PL-1**: Ganna Tolstanova	
	16:05–16:30	■ **I-1**: Polpharma Biologics	
	16:30–16:45	Coffee break	
	16.45–17.45	■ Oral session (**O-1**–**O-4**) *The Deathly Battle for Homeostasis*	■ Oral session (**O-5**–**O-6**) *Environment of Things*
	17.45–18.00	Coffee break	
	18.00–18.50	■ **PL-2**: Marco Costantini	
	from 18.50	Outdoor game	
Day 2: Saturday	10.00–10.50	■ **PL-3**: Aleksandra Ziembińska-Buczyńska	
	10.50–11.20	■ **I-2**: Proteon Pharmaceuticals	
	11.20–11.30	Coffee break	
	11.30–12.30	■ Oral session (**O-7**–**O-9**) *50 Shades of Plants*	■ Oral session (**O-10**–**O-13**) *The Deathly Battle for Homeostasis*
	12.30–12.40	Coffee break	
	12.40–13.30	■ **PL-4**: Michał Gabruk	
	13.30–14.40	Lunch break	
	14.40–15.10	■ **I-3**: Bioton	
	15.10–16.00	Poster session 1 (**P-1** – **P-21**)	
	16.00–16.50	■ **PL-5**: Alicja Puścian	
	16.50–17.10	Coffee break	
	17.10–18.10	■ Oral session (**O-14**–**O-16**) *Industry Within Four Walls of Murein*	■ Oral session (**O-17**–**O-20**) *To Unfold What's Folded*
	from 21.30	Social event, *Głębokie Gardło, Bednarska 28/30, 00-321 Warsaw*	
Day 3: Sunday	10.00–11.00	■ Oral session (**O-21**–**O-24**) *To Unfold What's Folded*	■ Oral session (**O-25**–**O-27**) *50 Shades of Plants*
	11.00–11.10	Coffee break	
	11.10–12.00	■ **D-1**: Discussion Panel	
	12.00–13.00	Poster session 2 (**P-22**–**P-43**)	
	13.00–14.10	Lunch break	
	14.10–15.00	■ **PL-6**: Jacek Jemielity	
	15.00–15.40	■ Closing & Prizes	

■ – Room 9B ■ – Room 102B

PL – plenary lecture **I** – industry lecture **O** – oral presentation **P** – poster **D** – discussion panel

Agenda

Friday, 12 May 2023.

from 13:00 **Registration**

15:00–15:15 **Opening of the Conference**

15:15–16:05 **Plenary lecture**

PL-1 Gut microbiota and kidney stone disease: Focus on oxalate-degrading microbiota

Ganna Tolstanova, *Taras Shevchenko National University of Kyiv (UA)*

16:05–16:30 **Industry lecture**

I-1 Upstream Process for Biosimilar manufacturing

Tomasz Pawłowski *Polpharma Biologics, Duchnice–Warsaw (PL)*

16:30–16:45 **Coffee break**

16:45–17:45 **Oral session: *The Deathly Battle for Homeostasis***

O-1 The novel mechanism of aggravating post-myocardial infarction heart failure during obesity - a molecular view

Jakub Tomaszewski, *Medical University of Warsaw (PL)*

O-2 Legionella pneumophila ceramides as an example of bacterial molecular mimicry

Bożena Kowalczyk, *Maria Curie-Sklodowska University, Lublin (PL)*

O-3 Clinical isolate characterization of Klebsiella pneumoniae antibiotic ciprofloxacin resistance in tertiary hospital Purwokerto, Central Java, Indonesia

Indah Sulistiyawati, *Jenderal Sedirman University, Purwokerto (ID)*

O-4 Neuroanatomical differences between individuals with early and late chronotypes

Patrycja Ściślewska, *University of Warsaw & Nencki Institute of Experimental Biology Polish Academy of Sciences, Warsaw (PL)*

16:45–17:45 **Oral session: *Environment of Things***

O-5 Vector role of juvenile stages of the common tick *Ixodes ricinus* for *Borrelia burgdorferi*, *Babesia microti*, *Rickettsia spp.*

Patrycja Łyzińska, *Faculty of Biology, University of Warsaw (PL)*

O-6 Polydopamine and Polypyrrole application in yeast-based biofuel cells: A short polymer synthesis for better function

Viktorija Reinikovaite, *State Research Institute Center for Physical Sciences and Technology, Vilnius (LT)*

17:45–18:00 **Coffee break**

18:00–18:50 **Plenary lecture**

PL-2 Digital manufacturing in biomedical research: a step towards engineering functional tissue and organ replicas *in vitro*

Marco Costantini, *Institute of Physical Chemistry, Polish Academy of Sciences, Warsaw (PL)*

from 18:50 **Outdoor game**

Saturday, 13 May 2023.

10:00–10:50 **Plenary lecture**

PL-3 Microcosm of possibilities

Aleksandra Ziembińska-Buczyńska, *Silesian University of Technology, Gliwice (PL)*

10:50–11:20 **Industry lecture**

I-2 Bacteriophage cocktails – a new sustainable solution for infectious agents

Elżbieta Fornal *Proteon Pharmaceuticals, Łódź (PL)*

11:20–11:30 **Coffee break**

11:30–12:30 **Oral session: *50 Shades of Plants***

O-7 Development of a plant expression system for the RBD domain of the Spike glycoprotein of the SARS-CoV-2 coronavirus

Zuzanna Czarnomska, *Medical University of Warsaw (PL)*

O-8 Biosynthesis of 3',4,6-trihydroxybenzophenone-2-O-β-D-glucopyranoside and decussatin-1-O-primeveroside in *Gentiana capitata* Buch.-Ham. ex D. Don embryogenic cell suspension culture

Michał Markowski, *Medical University of Warsaw (PL)*

O-9 Effect of copper ions on *in vitro* cultures of *Miscanthus × giganteus*

Gabriela Belniak, *Adam Mickiewicz University, Poznań (PL)*

11:30–12:30 **Oral session: *The Deathly Battle for Homeostasis***

O-10 Multi-Organ-on-Chip technology for reproduction of metastasis environment *in vitro*

Joanna Konopka, *Warsaw University of Technology (PL)*

O-11 Multi-Organ-on-Chip approach in cancer therapy

Paweł Romańczuk, *Warsaw University of Technology (PL)*

O-12 'No cell is an island' – the role of stromal-derived CD44 protein in the development of drug resistance in leukemia cells

Laura Turos-Korgul, *Nencki Institute of Experimental Biology, Polish Academy of Sciences, Warsaw (PL)*

O-13 Graphene oxide modulates integrins expression in cervical cancer

Kacper Kręgielewski, *Warsaw University of Life Sciences (PL)*

12:30–12:40 **Coffee break**

12:40–13:30 **Plenary lecture**

PL-4 Photocatalytic enzyme that re-shapes lipid membranes – linking sequence to the properties with neural networks

Michał Gabruk, *Jagiellonian University, Kraków (PL)*

13:30–14:40 **Lunch break**

14:40–15:10 **Industry lecture**

I-3 Biotechnology every day. From strain to final drug product

Dariusz Gutowski, Jeremy Launders, Edyta Wysocka
BIOTON SA, Macierzysz–Warsaw (PL)

15:10–16:00 **Poster session 1 (P-1–P-21)**

16:00–16:50 **Plenary lecture**

PL-05 How social groups shape individual behavior - neural underpinnings of social bonding

Alicja Puścian, *Nencki Institute of Experimental Biology, Polish Academy of Sciences, Warsaw (PL)*

16:50–17:10 **Coffee break**

17:10–18:10 **Oral session: *Industry Within Four Walls of Murein***

O-14 Application of bacterial cellulose as a excipient in tablets for oral drug delivery

Klaudia Snopek, *West Pomeranian University of Technology, Szczecin (PL)*

O-15 Droplet microfluidic system for passive selection and enrichment of bacteria producing biosurfactants

Klaudia Staśkiewicz, *University of Warsaw (PL)*

O-16 Seeking for a potent pyocyanin producer using statistical planning

Joanna Jabłońska, *West Pomeranian University of Technology, Szczecin (PL)*

17:10–18:10 **Oral session: *To Unfold What's Folded***

O-17 Modeling of a putative programmed cell death receptor from *Nostoc punctiforme*

Jędrzej Kubica, *University of Warsaw (PL)*

O-18 Kinetic Analysis of IFITs Interactions and the Role of IFITs complexes in IFIT1 Binding to IVT mRNA

Jingping Geng, *University of Warsaw (PL)*

O-19 Does PD-L1 signaling have an impact in the molecular change in classic Hodgkin lymphoma?

Hummaira Sadaf, *Maria Sklodowska-Curie National Research Institute of Oncology, Warsaw (PL)*

O-20 Mechanistic studies of nonenzymatic self-replication of alternative RNA forms using molecular dynamics

Barbara Lech, *Wroclaw University of Science and Technology (PL)*

from 21:30 **Social event**

Sunday, 14 May 2023.

10:00–11:00 **Oral session: *To Unfold What's Folded***

O-21 Avidity at its finest: Hsp70 and the art of efficient substrate delivery

Igor Grochowina, *University of Gdansk and Medical University of Gdansk (PL)*

O-22 Autoinhibition in class B JDPs: the key to efficient amyloid fibril disaggregation

Katarzyna Kalinowska, *University of Gdansk & Medical University of Gdansk (PL)*

O-23 An ancient tale of two JDPs: The evolutionary journey from protein folding to preventing Alzheimer's disease

Dominik Purzycki, *University of Gdansk & Medical University of Gdansk (PL)*

O-24 The effect of selected point mutations in spinach violaxanthin de-epoxidase on the course of reaction and enzyme structure

Anna Juszczyk, *Jagiellonian University, Kraków (PL)*

10:00–11:00 **Oral session: *50 Shades of Plants***

O-25 Metabolism of reactive oxygen species in the traps of *Nepenthes x ventrata* during nitric oxide-stimulated digestion

Maciej Arkadiusz Piekarniak, *Warsaw University of Life Sciences (PL)*

O-26 Alterations in ROS metabolisms in the traps of *Nepenthes x ventrata* processing digestion stimulated by nitric oxide

Agnieszka Wal, *Warsaw University of Life Sciences (PL)*

O-27 Analysis of fatty acid profile in extracts from selected oil rich plant pomace used in *Yarrowia lipolytica* yeast cultures

Antoni Świergocki, *Warsaw University of Life Sciences (PL)*

11:00–11:10 **Coffee break**

11:10–12:00 **Discussion panel**

D-1 Biotech Challenges in Central Europe: Navigating Career Paths

12:00–13:00 **Poster session 2 (P-22–P-43)**

13:00–14:10 **Lunch break**

14:10–15:00 **Plenary lecture**

PL-6 mRNA technology for therapeutic applications

Jacek Jemielity, *University of Warsaw (PL)*

15:00–15:40 **Closing & Prizes**

Poster Session 1

Saturday, 13 May 2023, 15:10–16:00

P-1 Continuous research work: difficulties and challenges working with primary cell cultures derived from human cancer cells

Agne Reičiuniene, *Vilnius University, Vilnius (LT)*

P-2 Three-dimensional (3D) model of ovarian cancer in Lab-on-a-Chip

Paulina Musolf, *Warsaw University of Technology (PL)*

P-3 *Reynoutria japonica* — invasive plant with the potential use in phytoremediation. Study in *in vitro* model

Alicja Matyjewicz, *University of Agriculture in Kraków (PL)*

P-4 Temporary immersion bioreactors and agitated cultures as the useful tool for synthesis biologically active phenolic compounds in *Reynoutria japonica* and its biologically active properties

Julia Sroka, *University of Agriculture in Kraków (PL)*

P-5 The nanostructure of the polymeric solution determines the delivery of macromolecules into cells via osmotic shock

Aneta Karpińska, *Institute of Physical Chemistry Polish Academy of Sciences, Warsaw (PL)*

P-6 Study of ATP bioluminescence metres in assessing decontamination of biotechnological laboratories

Oliwia Paszkiewicz, *West Pomeranian University of Technology, Szczecin (PL)*

P-7 Purification of air from mixture of volatile organic compounds by adsorption method

Martyna Jurkiewicz, *West Pomeranian University of Technology, Szczecin (PL)*

P-8 Antibiotic resistance of *Salmonella* sp. bacteria isolated from sewage sludge from the Silesina, Opole and Lower Silesian voivodenships

Daria Chrobak, *Polish Society of Bioinformatics and Data Science – BIODATA (PL)*

P-9 Exploring interactions between short RNA ended with synthetic cap analogs and an innate immune response protein IFIT1, using bio-layer interferometry technique

Jan Stadnicki, *University of Warsaw (PL)*

P-10 Changes in morphology and physiology of streptomycetes after contact with nanomaterials

Kamila Dubrowska, *West Pomeranian University of Technology, Szczecin (PL)*

P-11 Development of the method to lead immobilization in hydroponic cultivation of plants Using Ureolytic bacteria *Ochrobactrum* POC9

Aleksandra Goszcz, *University of Warsaw (PL)*

P-12 Analysis of the role of extracellular vesicles in the transmission of ferritin from human macrophages to cancer cells

Artur Wardaszka, *Prof. Waclaw Dabrowski Institute of Agricultural and Food Biotechnology – State Research Institute, Warsaw (PL)*

P-13 Analysis of the survivality of *Listeria monocytogenes* under bacteriophage pressure in digestate, produced in biogas plants

Artur Czajkowski, *Pomeranian Medical University, Szczecin (PL)*

P-14 Nitrogen cycling model in Arctic ornithogenic soil (Hornsund, Svalbard)

Julia Brzykcy, *Institute of Biochemistry and Biophysics, Polish Academy of Sciences, Warsaw (PL)*

P-15 Index patient in an alimentary tick-borne encephalitis (TBE) outbreak

Abdulla Hourani, *Medical University of Warsaw (PL)*

P-16 Evaluation of the biocidal activity of nanoparticles and their synergistic effects in the control of prototheca bovine mastitis

Magdalena Kot, *Warsaw University of Life Sciences (PL)*

P-17 Impact of copper nanoparticles and cisplatin on HeLa cervical cancer cells line and U251 glioblastoma cells line viability

Antonina Komosa, *Warsaw University of Life Sciences (PL)*

P-18 Lipolytic and proteolytic activity of antarctic bacteria

Zuzanna Niewiadomska, *Łukasiewicz Research Network – Institute of Industrial Chemistry, Warsaw (PL)*

P-19 When glial cells encounter hypoxia-ischemia – the involvement of autophagy

Paulina Gębala, *Mossakowski Medical Research Institute Polish Academy of Sciences, Warsaw (PL)*

P-20 A yeast-based system for studying interactions between human ACE2 and the Spike protein of SARS-CoV-2 coronavirus

Weronika Śliwińska, *Adam Mickiewicz University, Poznań (PL)*

P-21 Antioxidant properties of *Aralia racemosa* and *Aralia spinosa* (Araliaceae) methanolic extracts

Kajetan Biernacki, *Medical University of Warsaw (PL)*

Poster session 2

Sunday, 14 May 2023, 12.00–13.00

P-22 The effect of the lack of dystrophin and its individual isoforms on the proliferation and differentiation of the SH-SY5Y human neuroblastoma cell line

Martyna Makowska, *Adam Mickiewicz University, Poznan (PL)*

P-23 Identification of novel inhibitors of the main protease of SARS-CoV-2 using a yeast-based system

Wojciech Grabiński, *Adam Mickiewicz University, Poznań (PL)*

P-24 Evaluation of the effectiveness of bacteriophages in the *Staphylococcus aureus* and *Pseudomonas aeruginosa* bi-species biofilm

Dominika Miłek, *Pomeranian Medical University, Szczecin (PL)*

P-25 Biological activity of secondary metabolites extracted from plants belonging to genus *Helleborus* spp.

Marta Sobolewska, *University of Gdańsk & Medical University of Gdańsk (PL)*

P-26 Antioxidant properties of *Polyscias filicifolia* (C. Moore ex E. Fourn.) L. H. Bailey (Araliaceae) callus extracts

Joanna Szatko, *Medical University of Warsaw (PL)*

P-27 The nanofibrous mats for human cardiac cells culture

Zuzanna Iwoń, *Warsaw University of Technology (PL)*

P-28 Antarctic bacteria as a source of new compounds against anti-multidrug resistant microorganisms

Piotr Lorek, *Łukasiewicz Research Network -– Institute of Industrial Chemistry, Warsaw (PL)*

P-29 Analysis of the effectiveness of anti-staphylococcal bacteriophage therapy in a human blood model

Patrycja Olszewska, *Pomeranian Medical University, Szczecin (PL)*

P-30 *DNAJC30* gene variation in patients with Leber hereditary optic neuropathy

Aleksandra Maciejczuk, *University of Warsaw (PL)*

P-31 Evaluation of petroleum products biodegradation capabilities of *Pseudomonas* strains immobilized in alginate beads generated in microfluidic devices

Julia Karbowska, *University of Warsaw (PL)*

P-32 Effect of kappa opioid receptor activation on tumor cell proliferation in the mouse colon

Mikołaj Mierzejewski, *Medical University of Łódź (PL)*

P-33 Nanofibrous mats integrated with PC-PDMS-PC microsystem for hypoxia studies in cardiac cells

Dominik Kołodziejek, *Warsaw University of Technology (PL)*

P-34 Synthesis, modification and application of magnetic nanoparticles in phototermal therapy

Aleksandra Wolińska, *Warsaw University of Technology (PL)*

P-35 MLK4 mutations in metastasis

Vi Truong, *IMol, Polish Academy of Sciences, Warsaw (PL)*

P-36 Application of modern biotechnological, phytochemical and molecular techniques to obtain secondary metabolites from *in vitro* cultures of *Rindera graeca* (A. DC.) Boiss. Heldr. (Boraginaceae) transgenic roots

Patryk Zakrzewski, *Medical University of Warsaw (PL)*

P-37 Effect of locomotor exercise on the density of Vesicular Glutamate Transporter 2 distributed in the ventral horn of the lumbar spinal cord in rats with spinal cord injury

Karolina Godlewska, *Warsaw University of Life Sciences & Nencki Institute of Experimental Biology PAS, Warsaw (PL)*

P-38 Targeting PD-1/PD-L1 immune checkpoint, different profiles of druggability between human and mouse PD-L1

Justyna Kocik-Król, *Jagiellonian University, Kraków (PL)*

P-39 Characterization and potential applications of *Yersinia enterocolitica* phages in minced meat

Marta Gliźniewicz, *Pomeranian Medical University, Szczecin (PL)*

P-40 Study of the effect of selected saponins on the transport of cytostatic drugs into cells

Aleksandra Kowalska, *Warsaw University of Technology(PL)*

P-41 Comparison of antimicrobial properties of selected dairy cow udder care preparations based on propolis extracts

Weronika Jabłońska, *Warsaw University of Life Sciences (PL)*

P-42 The optimization of the immobilization process of bacteriophage T4 on a carrier based on bacterial cellulose

Wiktoria Świeboda, *West Pomeranian University of Technology, Szczecin (PL)*

P-43 Survival analysis of *Tenebrio Molitor* L. Beetles injected with neuropeptides under thermal stress conditions

Olga Blauth, *Adam Mickiewicz University, Poznań (PL)*

PL-1: Gut microbiota and kidney stone disease: Focus on oxalate-degrading microbiota

*Ganna Tolstanova**

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The number of bacteria in human body is the same order as the number of human cells(10^{13}), and their total mass is about 0.2 kg. The highest bacteria number is in the gut. The gut microbiota is exclusively responsible for several metabolic important functions, including vitamin and short chain fatty acid (SCFAs) production, amino acid synthesis, bile acid biotransformation, hydrolysis and fermentation of non-digestible substrates. It is widely acknowledged that the gut microbiota can be altered by a range of factors, such as antibiotic use, diet, stress, and infections. When a balanced interaction between the gastrointestinal tract and the resident microbiota is disrupted, intestinal and extraintestinal diseases may develop. To date, hyperoxaluria is considered the main risk factor for the formation of oxalate-calcium stones, which account for 75% of all kidney stones. Oxalate is a potentially toxic anion of dicarboxylic acid ($C_2O_4H_2$), which is widespread in both plants and mammals Despite the potential toxicity, humans lack the oxalate metabolizing enzymes and are not able to degrade excessive oxalate levels. The gut oxalate-degrading bacteria (ODB) play an important role in oxalate destruction expressing the catabolic enzymes formyl-CoA transferase (Frc) and oxalyl-CoA decarboxylase (Oxc). We will talk on the dichotomy between total fecal oxalate-degrading activity (ODA) and the ODB number in oxalate homeostasis. Moreover we will present a new perspective on the translational relevance of ODA assessing in the prediction of nephrolithiasis formation and personalized prescribing of probiotics or synbiotics.

Professor Ganna Tolstanova is a highly accomplished physiologist and professor of biology, with expertise in studying the barrier function of the intestine in normal and pathological states. She obtained her PhD and DSc degrees from the Faculty of Biology at Taras Shevchenko National University of Kyiv, Ukraine, and has since received numerous grants for her research. Professor Tolstanova is an Associate Editor for Frontiers in Pharmacology and a member of the Editorial Board for Ukr. Biochem J. She is also co-chair of the Basic Gastroenterology Unit at the Ukrainian Association of Gastroenterologists. With over 100 academic publications and several supervised dissertations, Professor Tolstanova is a respected and influential figure in her field. Her research aims to identify agents for correcting intestinal barrier function, and her work has contributed significantly to our understanding of this complex area of physiology. In addition to her impressive research credentials, Professor Ganna Tolstanova also holds a leadership role at Taras Shevchenko National University of Kyiv, where she serves as the Vice-rector for Research.

PL-2: Digital manufacturing in biomedical research: a step towards engineering functional tissue and organ replicas *in vitro*

*Marco Constantini**

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Nowadays, biomedical research is facing a series of significant problems. These include i) scientific challenges – the available in vitro and animal models are often inadequate for the recapitulation of human biology, ii) economic issues – the cost related to drug development is skyrocketing, and iii) ethical concerns – the use of animals for research purposes is perceived by the public opinion as cruel and unjust. In this context, digital manufacturing technologies represent one of the most promising solutions to address these limitations, enabling for a rapid, highly-customized manufacturing of cellularized constructs that can efficiently mimic both the structural and functional properties of native tissues/organs. During the lecture, we will give a brief overview of the state-of-the-art in musculoskeletal tissue engineering and talk about the upcoming innovations required to shake up this area of study.

Marco Costantini PhD DSc is a researcher at the Institute of Physical Chemistry, Polish Academy of Sciences. He holds a master's degree in industrial chemistry and a Ph.D. in chemical and processing engineering, both from La Sapienza University of Rome. Currently, he is leading a multidisciplinary team working at the convergence of materials science, biology, and microfluidics at the Institute of Physical Chemistry – Polish Academy of Sciences. His current research interests span from the development of digital manufacturing strategies for the synthesis of functionally graded porous materials with enhanced physicochemical properties to the invention of advanced strategies for in vitro modelling and in vivo repair of musculoskeletal system.

PL-3: Microcosm of possibilities

*Aleksandra Ziembicka-Buczyńska**

Silesian University of Technology, Gliwice, Poland

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Biotechnologies are developed on the basis of the nature's observation. We mimic natural processes in the laboratory and develop them for its usage at larger scale. Many biotechnologies, mainly in the field of environmental biotechnology, are microbe-based. Why? Because microbes are everywhere. They live in complex communities in which every single microorganism has its own unique role. Their metabolism is extremely flexible and they can adapt to almost any sort of conditions. We usually treat them as our enemies, usually thinking about the illnesses they cause. And it is so unfair! Why? Because most of the environmental microorganisms are biotechnological superheroes - cleaning water, soil, air, producing useful chemical compounds, generating green energy and even healing the building materials. It is real microcosm of possibilities for biotechnology.

Aleksandra Ziembińska-Buczyńska, PhD DSc prof. SUT – microbiologist, academic teacher, science communicator from the Environmental Biotechnology Department, Faculty of Power and Environmental Engineering of the Silesian University of Technology. She works in the field of the biotechnological potential of bacterial communities in the natural and technological environment researched using molecular biology and classical microbiology approach. Her scientific interests include the usage of microorganisms in environmental bioremediation and biotechnological production, including industrially relevant bioproducts, green energy production and circular economy. Director of the Science Popularization Center SUT. Member of the Association of the Spokesmen of Science and Council for the Science Dissemination of the Polish Academy of Sciences. Awarded in FameLab Poland, as Popularizer of Science 2018 (the award of PAP Science in Poland and Polish Ministry of Education and Science) and with POPScience Award of Silesian Science Festival.

PL-4: Photocatalytic enzyme that re-shapes lipid membranes – linking sequence to the properties with neural networks

*Michał Gabruk**

Department of Plant Physiology and Biochemistry, Faculty of Biochemistry, Biophysics and Biotechnology, Jagiellonian University, Kraków, Poland

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Light-dependent protochlorophyllide oxidoreductase (LPOR) is a unique example of an enzyme with a light-driven catalysis that was recently shown to re-shape lipid membranes into cubic or filamentous phase, depending on the presence of reagents. The enzyme is involved in the penultimate reaction of the chlorophyll biosynthetic pathway, has two substrates, two products and can be found in all oxygenic phototrophs, including bacteria, algae and land plants. In all of these organisms LPOR is a central actor that integrates multiple regulatory signals to finetune the rate of chlorophyll biosynthesis for optimal growth. The photocatalytic mechanism of the reaction allows to characterize the reagents binding with unprecedented precision with the use of low-temperature fluorescence measurements, molecular biology techniques and cryo-electron microscopy. At the same time, multiple variants of the enzyme have been found in different species that reflect a complicated evolutionary history of land plants. In this talk I will summarize our efforts to characterize the landscape of properties of LPOR enzyme and our attempts to link the sequence of LPOR variant to the properties with neural networks and state-of-the art microscopy techniques. Our results show how evolutionary processes can affect the properties of the enzymes leading to the emergence of new functions, growth of complexity and better adaptation of living organisms to new ecological niches.

Michal Gabruk PhD is a biochemist and assistant professor at Jagiellonian University working on the regulatory network of chlorophyll biosynthesis in plants. He integrates molecular biology, state-of-the art cryo-election microscopy techniques and computational approach to predict properties of the enzymes out of the sequence and model the cellular processes. His scientific interests include photocatalysis, evolution of novel functions of enzymes and astrobiology.

PL-5: How social groups shape individual behavior – neural underpinnings of social bonding

*Alicja Puścian**

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A great majority of the research on social behavior, including on influences of others on the choices made by an individual, has been done in human subjects. However, studying the brain underpinnings of those phenomena in humans poses significant limitations of the ethical and methodological nature. Thus, in the lecture I will illustrate how research on these topics can be performed using laboratory mice, a species considered a relevant model of mammalian social behavior. Although the neural background of sociability is less complex in mice than in humans, its key components are highly evolutionarily-conserved. A body of evidence shows that rodents display sophisticated forms of social behavior including helping or even consoling a partner after averse experiences. At the same time, the availability of genetically modified strains of mice, alongside the rich toolbox of genetic engineering methods, allows for advanced in-vivo studies of changing neuronal activity in behaving animals. Indeed, one can also manipulate the activity of the identified neuronal circuits in real-time, to evoke a behavior change. Taken together, I will show how applying multidisciplinary experimental protocols enables studying social behavior with all potency of contemporary neuroscience. The illustrated approach is a critical step on the research path aiming at discovering specific brain mechanisms underlying social influences on behavior.

Alicja Puścian PhD is a behavioral neurobiologist who focuses on the relationship between the brain and behavior, particularly in the context of emotions, motivation, and social interactions, utilizing knowledge from psychology, biology, and behavioral analysis. She is currently involved in the BRAINCITY project, which investigates how social bonds are encoded in the brain. During her doctoral studies at the Nencki Institute of the Polish Academy of Sciences, she designed and patented the Eco-HAB system with her colleagues, which is used for the automated assessment of social behavior in laboratory animals under semi-natural conditions. Eco-HAB, which was created based on knowledge from electronics, advanced computational methods, and ecology, is currently being used in many laboratories in Europe and North America. Her scientific work on the role of neuronal plasticity in behavior and its deficits observed in disorders such as autism and depression has been published in leading scientific journals, including Molecular Psychiatry, Cell Reports, and eLife. She is a recipient of the Young Scientist of the Year awards from the Polish Society for Neuroscience and the European Brain and Behavior Society, as well as the Fulbright STEM Impact Award.

PL-6: mRNA technology for therapeutic applications

*Jacek Jemielity**

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University of Warsaw, Poland

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For several decades, scientists from all over the world have been trying to discover effective methods of combating diseases that are difficult to treat with traditional methods, such as cancer, genetic rare diseases, and the last years in every aspect of our lives has been dominated by the pandemic caused by the coronavirus SARS-CoV-2. The hope for improving this situation is the so-called gene therapy, in which a therapeutic is delivered in the form of a genetic recipe, which is then expressed in the cells of the patient. In recent years, messenger RNA (mRNA), which is the genetic recipe for a specific protein, has received a great deal of attention in this context. A kind of culmination of these efforts was the development of mRNA vaccines against coronavirus, which were the first to be approved for widespread use. On the way to effective mRNA-based therapies, there have been a number of problems that have been solved, but there is also room for improvement. During the lecture, the speaker will present the idea of gene therapies and their enormous potential beyond anticancer and antiviral therapies. He will talk about the main problems associated with the development of this novel therapy and ways to solve them using biological and chemical methods, including those developed at the University of Warsaw.

Professor Jacek Jemielity, a chemist specialized in bioorganic chemistry and biochemistry of nucleic acids, is the head of the Laboratory of Bioorganic Chemistry at the Center for New Technologies, University of Warsaw, and the co-founder of ExploRNA Therapeutics, a UW spin off company focused on developing mRNA modification technologies and developing innovative mRNA-based therapies. He is involved in research on the synthesis, properties, and applications of chemically modified nucleotides. He develops methods to synthesize biologically important nucleotides, creates tools to modify nucleic acids useful in studies of genetic information expression and medical applications. He developed a method to obtain mRNA with properties necessary for therapeutic applications. His inventions are used in several clinical trials on cancer immunotherapy. One of these technologies involving mRNA modification is used by leaders in biotechnology industry companies such as BioNTech, Sanofi, Roche, Pfizer in clinical trials for cancer vaccines. He is a winner of the President of Poland's Economic Award in the "research + development" category (2017), and was nominated in the competition organized by the European Patent Office "European Inventor Award" 2018 in the category "Research". Recently, in 2021, he also received the Foundation for Polish Science Prize in chemistry and materials sciences for developing chemical modifications of mRNA as tools for therapeutic applications, and studies on cellular processes.

I-1: Upstream Process for Biosimilar manufacturing

*Tomasz Pawłowski**

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If you're interested in learning about the innovative world of biotechnology and biosimilar manufacturing, don't miss out on our upcoming lecture! We will be sharing valuable insights on the upstream process for biosimilar manufacturing, which is critical for the development and production of these life-changing medicines. That is why in Polpharma Biologics we are highly focused on advancing and expanding our pipelines to increase the patient access to these much-needed biosimilar drugs. We will dive into the latest innovations and techniques used to produce high-quality biosimilars, providing you with a comprehensive overview of the industry. Whether you're a student, researcher, or simply curious about biotechnology, this lecture is the perfect opportunity to expand your knowledge and network with like-minded individuals.

Polpharma Biologics is a biotechnology group active in the field of development and manufacturing of biologics. The first Polish company to manufacture a biosimilar drug that has been approved both by the FDA and EMA. Polpharma Biologics has been active for more than 10 years pursuing R&D operations on a scale unprecedented in the Polish biotechnology sector. The group has been consistently investing not only in laboratories and production lines, but also in building the know-how to create highly specialized job opportunities and forging closer bonds of cooperation between the scientific and business communities.

I-2: Bacteriophage cocktails – a new sustainable solution for infectious agents

Elżbieta Fornal, Jolanta Witaszewska, Agnieszka Maszewska, Joanna Kazimierczak, Ewelina Wójcik, Jarosław Dastych*

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Proteon Pharmaceuticals was founded to implement a unique mission: to build a technology platform for isolation, characterization and production of bacteriophage cocktails, in order to replace the antibiotic use and reduce antimicrobial resistance of bacterial pathogens. Bacteriophages were first successfully used to treat bacterial infections a decade before penicillin was discovered. They are perfectly natural bacterial antagonists, specific only towards their host strains and neutral for the remaining members of the microbiome. For the first few decades bacteriophages were severely underappreciated. Only now, in the face of an overwhelming threat from antimicrobial resistant strains, their unique mode of action, restricted to the pathogen and not the patient, has become a quickly developing method of treatment. The exquisite features of bacteriophages, so opposite to antibiotics, are being explored. Unlike artificial chemicals, phages are nontoxic, completely biodegradable and the phage therapy triggers no side effects. Our main goal is to draw from the benefits of bacteriophages' existence to increase the food safety, raise the bar on control of pathogens and most importantly improve our quality of life. The technology platform created by Proteon Pharmaceuticals allows to successfully develop phage-based products and implement new strategies of control of pathogenic bacteria affecting animal production industry. The platform allows to design highly effective products consisting of a mixture of carefully selected and genotypically characterized virulent phages that eliminate pathogenic bacteria, without causing side effects, while supporting better performance outcomes. Proteon is a company oriented on sustainability. Our processes are optimized to minimise negative environmental impacts, conserve energy and natural resources, are safe for employees, communities and consumers. Our first Impact Report is available since May 8th, showcasing our contribution to a more sustainable food systems by reducing the unnecessary use of antibiotics, supporting food safety, increasing sustainability in food production, reducing food waste and ensuring food security.

Proteon Pharmaceuticals is a leader in bacteriophage technology for livestock farming and aquaculture. They help eliminate the unnecessary use of antibiotics from the food chain to reduce the risk of antimicrobial resistance (AMR), increase environmental sustainability and enhance animal welfare in protein production. Their patented phage technological platform combines substantial developments in genomics, bioinformatics, AI, and molecular biology to deliver precision feed additives that are effective, reliable, and safe. Proteon is also hiring talented people to join them in their mission. Proteon pharmaceuticals' Team combines experts with recent graduates from diverse backgrounds. They are global in our outlook and culture. All of the teams, including Bioinformatics, R&D, Product Development, and Manufacturing, are constantly growing, and you could be a part of Proteon's future. Join the Team to experience working in cutting-edge labs with a friendly work environment and great professional opportunities.

I-3: Biotechnology every day. From strain to final drug product

*Dariusz Gutowski**, *Jeremy Launders*, *Edyta Wysocka*

BIOTON SA, Macierzysz–Warsaw, Poland

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During the lecture, you will learn how the industry uses biotechnology to produce life-saving drugs. We will discuss the requirements for the preparation of production strains and the establishment of cell banks. Together we will go through the full biotechnological process of active substance production, formulation, sterile filtration and final sterile dosing and packaging. You will learn how to transfer technologies from the laboratory scale to the production line, based on cGMP requirements. Finally, a portfolio of analytical methods will be presented that confirm the highest quality of manufactured drugs.

BIOTON: 1 of 8 global producers of recombinant human insulin (RHI) and the only one in Poland producing insulin from the biosynthesis of the active substance to the finished product full cycle of insulin production. Production of human insulin is based on E. coli bacteria – their DNA was modified by our scientists to provide a safe, predictable and extremely efficient way of production. Facility GMP-compliant and approved in European Union. Quality Control and Quality Assurance designed to meet EMA requirements.

Pfizer: Breakthroughs that change patients' lives. Pfizer harnesses scientific knowledge and global resources to provide patients with therapies that extend their lives and dramatically improve their quality of life. We are committed to setting the standard for quality, safety and value in the discovery, development and manufacture of innovative medicines and vaccines. Every day, Pfizer employees promote health and advance preventative and curative treatments and therapies that address some of the most dangerous diseases of our time. As one of the world's leading, innovative biopharmaceutical companies, we have a strong responsibility to partner with healthcare providers, governments and local communities to increase the availability of high-quality, affordable healthcare around the world. For more than 170 years, we have been helping those who count on us.

D-1: Biotech Challenges in Central Europe: Navigating Career Paths

In this panel discussion we will shed light on the current landscape of the biotech industry, explore career opportunities, and discuss strategies for success. Our panelists will address key questions and topics such as: (1) current challenges faced by the biotech industry in Central Europe, (2) choosing the best career path in the biotech industry: factors to consider and tips for success, (3) essential skills required to excel in the biotech industry, (4) major trends shaping the biotech industry and their impact on Central Europe, (5) encouraging young talents to pursue careers in biotechnology.

By participating in this panel discussion, you will gain valuable knowledge, broaden your understanding of the biotech industry's challenges and opportunities, and receive practical insights to navigate your career path effectively. Join us to connect with industry experts, network with like-minded professionals, and contribute to the growth of the biotech industry in Central Europe. Don't miss this exciting opportunity to be part of the conversation!

What can you expect from our panelists?

Małgorzata Masierek, Medical Marketing Director at Bioton, will discuss challenges faced by biotech companies, strategies for attracting and retaining top talents, exciting innovations in the region.

Magdalena Kulczycka, the Director of Central European BioForum, will share her experiences as a molecular biologist, entrepreneur, and technology broker. She will provide insights into the evolution of the biotech industry in Central Europe, opportunities for professionals and future bioetchnology graduates.

Agnieszka Gawda, Vice-President and CBO at Polonium Foundation, will shed light on the integration of the Polish scientific community, she will also discuss the ways in which the biotech industry in Central Europe can better support its expat workforce.

The discussion will be moderated by **Marcin Szymon Filipiak**, co-founder and former president of WSB "Symbioza".

O-1: The novel mechanism of aggravating post-myocardial infarction heart failure during obesity – a molecular view

Jakub Tomaszewski[*,1], *Katarzyna Czarzasta*[1], *Karol Momot*[1], *Małgorzata Wojciechowska*[1]

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Dietary fat overload and obesity promote cardiac remodeling leading to unfavorable outcomes after myocardial infarction (MI). Recent studies on a mice model showed that nitrosative stress (NS) aggravates heart failure (HF) with preserved ejection fraction. NS consists of two important elements: the overexpression of inducible nitric oxide synthase (iNOS) and the inefficient unfolded protein response (UPR) pathway – particularly the XBP1s-IRE1α branch. We examined the impact of high fat diet (HFD) on the myocardial NS after MI-HF on an animal model. We performed the study on 29 male adult Sprague Dawley rats. We assigned them to groups and fed them for 8 weeks with normal diet (NFD) and HFD. Then, on the 12th week of age, we performed surgical procedures: evoking of MI (HF NFD, HF HFD) and sham operation (SO NFD, SO HFD). 4 weeks past the procedures, we sacrificed the animals, collected heart tissue and blood for further analysis. We confirmed HF development measuring NT-proBNP concentration in blood by ELISA and evaluate the levels of the following proteins in the heart tissue: IRE1α, phosphorylated IRE1α (pIRE1α), iNOS and β-actin as a control by Western blot. Both HF HFD and HF NFD groups developed post-MI HF according to higher plasma NT-proBNP concentration. The HF HFD group presented the highest expression of iNOS of all groups. The results correspondence to the development of NS in this group. Our data imply that the combination of HFD and MI generates the most severe MI-HF compared to isolated HFD and MI. The ratio of pIRE1α to IRE1α represents the activity of the UPR pathway. The IRE1α activity was downregulated in HF-HFD, but not in the HF-NFD. The data suggest that HFD significantly downregulates the UPR pathway after MI-HF. Our findings suggest that MI increases NS and downregulates the UPR pathway in HFD rats, but not NFD. This is a new perspective in future studies – the NS and UPR pathway as a therapeutical target in post-MI HF development.

Keywords: Heart Failure, Obesity, Unfolded Protein Response Pathway

O-2: *Legionella pneumophila* ceramides as an example of bacterial molecular mimicry

Bożena Kowalczyk[*,1], *Jacek Tarasiuk*[1], *Beate Fuchs*[2], *Christina E. Galuska*[2], *Katarzyna Pastuszak*[3], *Małgorzata Jurak*[1], *Marta Palusińska-Szysz*[1]

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[2] Core Facility Metabolomics, Research Institute for Farm Animal Biology (FBN), Dummerstorf, Germany
[3] Department of Interfacial Phenomena, Maria Curie-Sklodowska University, Lublin, Poland

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Legionella represents a narrow group of microorganisms that have adapted to the intracellular lifestyle. Inhalation of aerosol containing the bacteria can result in severe pneumonia in humans, called Legionnaire's disease. Once *L. pneumophila* invades the human lungs, the bacteria replicate within alveolar macrophages. *L.pneumophila* has developed an array of mechanisms that allow the bacteria to enter, survive and multiply in host cells. One of those mechanisms is molecular mimicry, exemplified by synthesizing ceramides structurally similar to ceramide produced by mammalian cells. The aim of this study was the investigation of *L. pneumophila* lipid composition, especially ceramides. Lipids were extracted from the bacteria by Bligh and Dyer method and analyzed by ultra-performance liquid chromatography coupled to tandem mass spectrometry. Mass spectrometry-based lipidomic analysis showed a complex lipid composition that included both neutral lipids (triglycerides, diglycerides, ceramides) and phospholipids (phosphatidylcholine – PC, phosphatidylethanolamine – PE, phosphatidylglycerol – PG, cardiolipin – CL). The most abundant lipid fractions were PC and bis-methyl phosphatidylethanolamine. The ceramides comprised approximately 0.02% of the total relative content of detected lipids. *L. pneumophila* synthesized 10 various ceramide species and one oxidized ceramide (t20:0_24:0+O). The dominant species of ceramides were cer(d37:1) (13.5%), cer(t18:0_26:0) (13.2%), cer(t35:0) (12.1%) and cer(t20:0_26:0) (11.0%). The structure of *L. pneumophila* ceramides is similar to that of mammalian ceramides, whose acyl chain lengths range from 14 to 26 carbon atoms with palmitic (C16:0) and stearic (C18:0) acids as the most common fatty acids. Determining the complex composition of bacterial membranes may help to unravel the pathogenicity mechanisms of *L. pneumophila* and its development toward antibiotic resistance.

The study was supported by the grant NCN 2017/27/B/NZ6/01544.

Keywords: *Legionella pneumophila*, Legionnaire's disease, molecular mimicry, ceramides, lipidomic analysis

O-3: Clinical isolate characterization of *Klebsiella pneumoniae* antibiotic ciprofloxacin resistance in tertiary hospital Purwokerto, Central Java, Indonesia

Indah Sulistiyawati[*,1], *Daniel Joko Wahyono*[1], *Wahyu Siswandari*[2]

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Ciprofloxacin is a broad-spectrum and effective quinolone derivative antibiotic used today, showing a high pattern of resistance in several countries. *Klebsiella pneumoniae* is the main human pathogen causing cases of infection in recent decades. His presence in the hospital is a major concern because of the high cases of resistance to antibiotics. This study aims to characterize *Klebsiella pneumoniae* in several clinical specimens of patients at Prof. Hospital. Dr. Margono Soekardjo Purwokerto Indonesia. This research has received approval from the Ethics Commission Number: 420/08640. In this study isolates of ciprofloxacin resistant *K. pneumoniae* were obtained from clinical specimens of blood, sputum, urine, pus, feces, pleural fluid during August-October 2022. Identification and sensitivity test of *K. pneumoniae* to ciprofloxacin used Vitek 2 ® Compact. The results showed that 155 *K. pneumoniae* isolates, with the majority of the patients being 86 (55.48%) male and 69 (44.52%) female. Sample specimens were obtained from patients who were treated in ICU, PICCU, ICCU, HCU, Surgery Poly, inpatient care. With the majority coming from the age group of patients >60 years (56.77%). Based on the qualifications of the collected specimens, blood samples were obtained (10/ 6.45%), sputum (101/ 65.16%), pus (27/ 17.42%), urine (13/ 8.39%), feces (3/ 1.94%) and pleural fluid (1/ 0.65%). Testing the sensitivity of *K. pneumoniae* ciprofloxacin isolates obtained 72 (46.5%) resistant isolates, 73 (47%) sensitive and 10 (6.5%) intermediate isolates. The prevalence value included in the calculation formula for cases of ciproflocxin resistance during August-October 2022 is 46.5%. In conclusion, ciprofloxacin resistant *K. pneumoniae* subspecies pneumoniae was found in several clinical specimen samples, indicating the need for renewal of management in administering antibiotics.

Keywords: *K. pneumoniae* , ciprofloxacin, resistant, clinical specimens

O-4: Neuroanatomical differences between individuals with early and late chronotypes

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The internal biological clock and the endogenous chronotype are essential, though often overlooked, variables in neurobiological research. Diurnal time preferences allow us to define the phase of the circadian rhythm. However, this timing is greatly varied, making the individual chronotypes fall on a continuum, at both ends of which, there are groups of early (EC) and late (LC) chronotypes. The aim of the analyses was to compare the neuroanatomy of people with extreme chronotypes. It has been hypothesized that LC people are more likely to experience sleep deprivation and its long-term effects – changes in brain anatomy. To test the hypothesis, the MRI T1 scans of 136 people were analyzed using MATLAB and SPM12, CAT12, xjview. Chronotype and sleep characteristics were determined using MEQ and PSQI questionnaires. Statistical analyses were based on two samples t-test and ANCOVA. Brain volume, age and sex were used as covariates. VBM analysis (voxel-level $p < 0.001$, cluster-level FWE < 0.05) showed 3 enlarged areas in EC brains (compared to LC) – Left Lingual Gyrus (Z-score = 3.67), Parahippocampal Gyrus (Z-score = 3.52) and Left Cerebellum (Z-score = 3.15), with no statistically significant differences in the whole brain volume of EC / LC (1513.74 ± 139.89 cm^3 and 1503.28 ± 131.31 cm^3, respectively). The results indicate significant neuroanatomical differences, which broaden our knowledge about chronotypes, but it is difficult to define the chain of cause and effect, so longitudinal studies are needed. Further research will investigate potential differences between people with different amplitudes of circadian rhythm – the second key characteristic of chronotype.

Acknowledgment: This work was supported by Ministry of Science and Higher Education (Poland) as a project under the program Excellence Initiative – Research University (2020–2026) no. BOB-IDUB-622-28/2023 (IV.4.1.). Data analyzed in the current project is now available online (Zareba et al., 2022).

Keywords: chronotype, MRI, neuroanatomy, biological clock

O-5: Vector role of juvenile stages of the common tick *Ixodes ricinus* for *Borrelia burgdorferi*, *Babesia microti*, *Rickettsia* spp.

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The common tick *Ixodes ricinus* is an ectoparasite of vertebrates, including humans. Based on current knowledge, adult *Ixodes ricinus* ticks carry many pathogens of medical and veterinary importance. A single tick can carry more than one pathogen, transmitting infectious diseases. One of the most common tick-borne diseases is Lyme disease, which is caused by bacteria - the spirochetes *Borrelia burgdorferi*. Babesiosis is another tick-borne disease, caused by the protozoan *Babesia microti*, which can occur simultaneously with Lyme disease. Dangerous pathogenic bacteria transmitted by *Ixodes ricinus* are also rickettsiae that cause many serious diseases belonging to the rickettsiosis group.

The purpose of my research is to test the possibility of transmission of pathogenic pathogens – *Borrelia burgdorferi*, *Babesia microti* and *Rickettsia* spp. by juvenile forms of the common tick *Ixodes ricinus*, larvae and nymphs.

In order to find an answer to the research question, *Ixodes ricinus* ticks were collected from vegetation in and around Urwitalt. The larvae were pooled 10 at a time and the nymphs were tested individually. DNA extraction of the ticks was performed and then molecular PCR technique was used to detect pathogen DNA using specific primers. PCR products were detected on agarose gel after electrophoresis. DNA of *Rickettsia* spp. was identified in 26%, *Borrelia burgdorferi* in 11% and *Babesia microti* in 1.6% of juvenile ticks. The research is still in progress, but already at this stage I have confirmed the presence and possibility of transmission of the pathogens *Borrelia burgdorferi*, *Babesia microti* and *Rickettsia* spp. by juvenile stages of the *Ixodes ricinus* tick.

Keywords: Vector role of juvenile stages of the common tick, *Ixodes ricinus*, *Borrelia burgdorferi*, *Babesia microti*, *Rickettsia* spp., larvae and nymphs

O-6: Polydopamine and Polypyrrole application in yeast-based biofuel cells: A short polymer synthesis for better function

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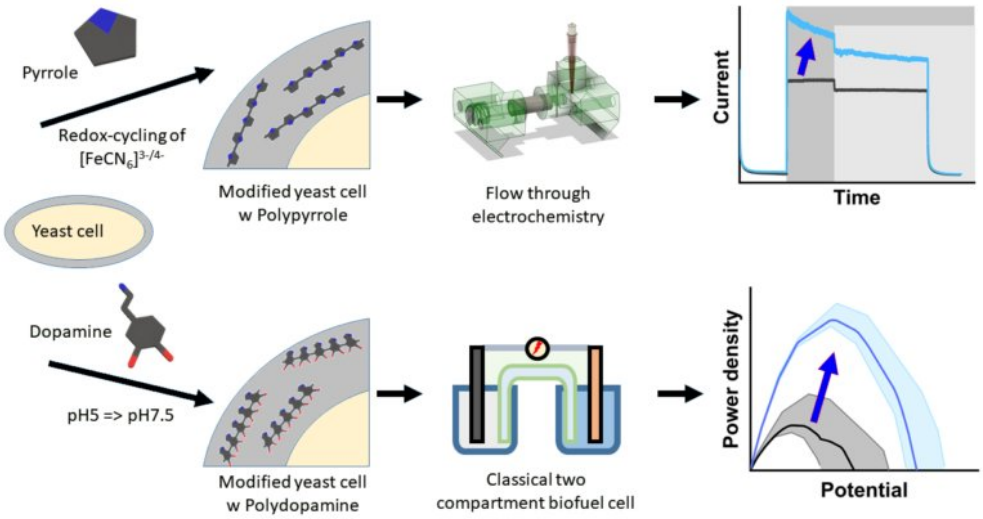
Microbial fuel cells (MFCs) is a new emerging technology capable of producing bioelectricity from organic materials through the metabolic processes taking place within the electroactive microorganism. However, low bioelectricity yield impedes MFC technology from further technological implementation. In this study we used yeast-based MFC, where two electricity-conducting polymers (polydopamine(PDA)and polypyrrole(Ppy)) were tested for improving charge transfer across the cell membrane/wall. Here we show that a short 2-hour modification significantly increased cell electrical properties, where PDA is more beneficial for MFC design, while Ppy is more suited for biosensor creation. We found that PDA modification can be easily controlled by varying the basicity of the incubation buffer from pH 5.0 to 7.5. Potentiometry experiments using different external resistances (10Ω–1MΩ) showed that: MFC based on PDA modification had a peak power value of 113 μW m^{-2} compared to control 50 μW m^{-2}, while Ppy-modified yielded 115 μW m^{-2} compared to control 84 μW m^{-2}. Chronoamperometric experiments in a dynamic flow-through system revealed that PDA modification yielded 35% higher current, whereas Ppy increased the current by 37.5% which dramatically decreased overtime.

Keywords: Microbial Fuel Cells, Conducting polymer, Polydopamine, Polypyrrole



O-7: Development of a plant expression system for the RBD domain of the Spike glycoprotein of the SARS-CoV-2 coronavirus

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Among the leading products of the biopharmaceutical industry, recombinant proteins occupy a very important position, being at the same time the cause of therapeutic progress in the field of oncology, infectious diseases and many others. Given the high cost of mammalian expression systems and the limitations specific to bacterial production, an efficient alternative would be economically beneficial to the pharmaceutical industry and the healthcare sector. Compared to these conventional methods, plant expression systems offer high throughput and scalability, elimination of animal-derived components from the production line and relative ease of adaptation to the principles of Good Manufacturing Practice, which makes them a highly competitive solution for the production of recombinant proteins. The presented research was aimed at expressing the recombinant vaccine antigen of the Spike glycoprotein of the Sars-CoV-2 coronavirus in a new plant expression system based on *in vitro* cultures of the *Nicotiana tabacum* BY-2 cell suspension. The originally designed plasmid carrying the expression cassette for the Spike RBD gene selected for the study, under the control of the CaMV 35S constitutive promoter, was obtained based on the pGFPGUSPlus vector. The resulting plasmid was introduced by electroporation of Agrobacterium tumefaciens strain LBA4404, which was then cultured on YMB selection medium containing kanamycin. Bacteria carrying the recombinant plasmid were used to agroinfect *N. tabacum* BY-2 cells. The resulting transformants were selected for 28 days on modified LS medium containing hygromycin and cefotaxime. Molecular analysis of restriction digest products and histochemical staining for β-glucuronidase activity were performed to confirm agroinfection. Qualitative proteomic analysis and confirmation of the production of the target protein were performed using the Western blot technique, after protein isolation by Ni-NTA affinity chromatography (IMAC).

Keywords: *Nicotiana tabacum* BY2, protein expression, recombinant vaccine antigen, Sars-CoV-2

O-8: Biosynthesis of 3',4,6-trihydroxybenzophenone-2-O-β-D-glucopyranoside and decussatin-1-O-primeveroside in *Gentiana capitata* Buch.-Ham. ex D. Don embryogenic cell suspension culture

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Plants occupies special role in pharmaceutical industry. Their secondary metabolites are important as a source of modern drugs, novel bioactive compounds and leading structures for drug development. Aim of the study was to obtain *Gentiana capitata in vitro* cell suspension cultures and identify their some secondary metabolites. *G. capitata* is a rare Himalayan plant species which is poorly studied in terms of biotechnology and phytochemicals. Seeds were obtained from the botanical garden and then sterilized, germinated on MS medium supplemented with GA3. Callus was induced from cotyledonary explants on MS medium supplemented with kinetin and 2,4-D. Next callus was cultured in liquid medium with the same composition on rotary shaker leading to plant suspension culture. Biomass obtained from suspension culture were lyophilized and extracted. Total extract were screened on TLC and fractionated by increasing polarity solvents. Two dominant compounds from mildly lipophilic and hydrophilic fractions were isolated by normal phase FLASH and reverse phase preparative HPLC. Structures of isolated compounds were elucidated by NMR and LC-MS. New compounds were screened for cytotoxicity by MTT assay on human cancerous, immortalized and primary cell lines A549, HaCat and CCD16Lu. In this study it was shown that *G. capitata* is a sustainable source of two new secondary metabolites 3',4,6-trihydroxybenzophenone-2-O-β-D-glucopyranoside and decussatin-1-O-primeveroside. These compounds show low toxicity to all cell lines that were tested. Viability in each cell line was greater than 80% in range 0-50 μg/ml. Further study should be focused on finding specific molecular target for these nontoxic compounds.

Keywords: *Gentiana capitata*, plant cell suspension, secondary metabolite isolation, MTT assay

O-9: Effect of copper ions on *in vitro* cultures of *Miscanthus × giganteus*

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In plant biotechnology, *in vitro* cultures is extremely important and widely used method, which allow the cultivation of plant cells, tissues or organs under strictly controlled, aseptic conditions. One of the paths in obtaining new plants due to the technique is indirect somatic embryogenesis, which results in the formation of somatic origin embryos. This method has many applications, including maintaining gene banks, micropropagation, obtaining transgenic plants or new varieties. However, *in vitro* cultures exhibit some limitations. For some species, long-term cultures of plant tissues turn out to be particularly problematic due to the low morphogenic potential, which decreases with the duration of the culture. *Miscanthus × giganteus* is one of the species characterized by short-term regenerative potential in callus cultures. In the study, in order to establish conditions for long-term culture of embryogenic callus, the MS medium was modified with the addition of $CuSO_4$. The substance shows positive effects on the maintenance of tissue cultures by, among other things, stimulating cells to produce antioxidants, including phenols. In the presented research, analyses were performed to determine soluble phenols in *M. giganteus* callus by spectrophotometric method. Further, for the purpose of understanding the effect of $CuSO_4$ on the regenerants obtained during somatic embryogenesis, the produced plants were subjected to multidisciplinary analyses, including measurements of photosynthetic gas exchange and RAPD-PCR (Random Amplified Polymorphic DNA Polymerase Chain Reaction).

Keywords: somatic embryogenesis, copper ions, long-term culture, *Miscanthus × giganteus*

O-10: Multi-Organ-on-Chip technology for reproduction of metastasis environment *in vitro*

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Malignant neoplasms and related metastases are one of the leading causes of death worldwide. Numerous scientific attempts to better understand the unknown phenomena associated with this disease have been made. However, both *in vitro* and animal models have proved to be inadequate. To better understand the processes taking place during the spread of neoplastic cells, new models able to faithfully reproduce tumor microenvironment, metastatic niche and tissues connecting them are desired. Some Organ-on-chip approaches have been proposed, combining microfluidic systems and small culture chambers that can faithfully model organs and their connections. However, standard platforms of this type do not go beyond the organ level. Moreover, they examine processes taking place during metastasis separately in the tumor, the metastatic niche, or the endothelium. Therefore, we propose a new research approach to understand cancer metastasis by developing a multi-Organ-on-Chip system platform using microflow and 3D cell models. Different geometries of microplatform were executed with the use of 3D printing technique. Final version of microsystem consists of two layers. The upper layer (PDMS) contains network of channels, while the bottom (microscope slide) acts as a sealing. Design of a microchip includes 5 parallel microchannels. Cylindrical microchannel seeded by Human Umbilical Vein Endothelial Cells (HUVECs) was generated by viscous finger patterning technique within central microchannel. Its diameter corresponds to the size of vessels smaller than human arteries (438.5 $\pm$ 10.1 μm). Lateral microchannels allowed the introduction of tumor spheroids, liver spheroids and cell medium. Spatial arrangement of introduced cells, their morphology and viability were visualized by fluorescence and confocal microscopy. Research conducted so far is the basis for developing desired multi-Organ-on-Chip platform.

This work was funded by BIOTECHMED-3 No. 504/04496/1020/45.010420.

Keywords: multi-organ-on-a-chip, breast cancer, metastasis, viscous finger patterning (VFP), microfluidics

O-11: Multi-Organ-on-Chip approach in cancer therapy

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Cancer is the leading cause of death worldwide, accounting for nearly 10 million deaths in 2020, where the most common was breast cancer. Currently, there are many methods of cancer treatment i.e. hormone therapy, immunotherapy, as well as the most used chemotherapy. Despite such widespread use of chemotherapy, it is associated with a high incidence of severe side effects. In our *in vitro* studies, we used a microsystem made of PDMS, which consisted of two chambers separated by a porous membrane made of polycarbonate for the liver and cancer model and one chamber for skin model. The chambers were connected to each other by a channel that allowed the delivery of a nutrient medium, a drug, as well as intercellular communication. The microsystem allows 3D formation of the liver and the cancer model, as well as skin model as a 2D monolayer. The liver and breast cancer model were created by composing the appropriate cell lines (liver - hepatocytes HepG2 and hepatic stellate cells HSC; breast cancer - tumor cell line MDA-MB231 and breast fibroblasts HMF) and placing them in collagen type I solution and then inserting them through separate channels into the lower chamber (under the membrane). The skin model, on the other hand, was created by introducing skin cells (keratinocytes HaCaT) to the surface of the chamber, which had previously been coated with collagen type I solution. Cell culture was carried out in the microsystem for 7 days. At 48h after model formation, a medium with the chemical compound in the form of the drug 5-fluorouracil, as well as the pro-drug capecitabine was administered. The proliferation and viability of each organ model was then checked using AlamarBlue® and claceinAM/propidium iodide, as well as at the end of the ongoing culture. Administration of the drug resulted in inhibition of proliferation in all cell models. Using HPLC-ESI-MS/MS it was confirmed that the liver model functions properly and degrades the pro-drug to its active metabolite.

Keywords: multi-organ-on-chip, cancer, microsystems

O-12: 'No cell is an island' – the role of stromal-derived CD44 protein in the development of drug resistance in leukemia cells

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The interactions between leukemia cells and their microenvironment play an important role in the development of drug resistance. Tunneling nanotubes (TNTs) represent a novel type of direct cell-cell communication way allowing for the direct transfer of different types of cargo between distant cells. Our previous studies have shown that these structures are formed between bone marrow stromal HS-5 cells and chronic myeloid leukemia (CML) K562 cells. Significantly, the TNT-mediated transfer of membrane vesicles from stromal to leukemic cells resulted in the stroma-mediated protection of CML cells from imatinib-induced apoptosis. Our previous trans-SILAC proteome studies have shown that together with vesicles, the functional groups of proteins with a potential role in protection and adaptation can be transferred from stromal to CML cells. Among them, CD44 – a protein associated with drug resistance, was found in acceptor cells. Therefore, we hypothesized that stromal cells support leukemic progression by direct transfer of CD44 towards leukemia. Presented studies aimed to confirm the TNT-mediated transfer of CD44 from stromal to CML cells and elucidate the role of stromal-derived CD44 protein in leukemia cells. We observed that after co-culture of CML cells with stroma, the level of CD44 in leukemia increases compared to cells grown in monoculture. Moreover, images obtained by confocal microscopy confirmed the presence of the stromal CD44-OFP protein on the cell membrane of leukemic cells. Flow cytometry analyses of rhodamine-123 efflux indicated significantly increased efflux pump activity in CML CD44+ cells. Our data show for the first time that bone marrow stromal cells can transfer CD44 protein to leukemia through direct cell contact, including TNTs. Moreover, CD44 presented in acceptor cells may be involved in the development of drug resistance in CML cells, by promoting the activity of efflux transporters.

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Keywords: leukemia, tunneling nanotubes, TNT, chemoresistance

O-13: Graphene oxide modulates integrins expression in cervical cancer

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Integrins are a family of membrane receptor proteins involved in the adhesion of cells to the extracellular matrix (ECM) and other cells. In cancer, integrins play role in cancer cell migration, proliferation, angiogenesis and tumor progression. Integrins are promising targets of selective cancer therapy because of the broad range of cellular functions that they control. Cervical cancer is the third most common type of cancer with which women are diagnosed. Due to the lack of adequate medical care, cervical cancer remains a common cause of death in underdeveloped countries. Graphene oxide (GO) is an oxidized derivative of graphene which is widely used in many industries, including cancer research.

We examined the effect of triple treatment with GO on cervical cancer HeLa and non-cancerous HS-5 cell lines on the expression of genes encoding proteins of the integrin family. GO was administered at 48h intervals at a concentration of 1 μg/mL. After 24h last treatment RNA was isolated and cDNA synthesized followed by qPCR to determine the expression of *itga3*, *itga5*, *itga6* and *itgav* genes. Analysis of gene expression showed that triple GO treatment resulted in decreased expression of *itga3*, *itga5*, *itga6* and *itgav* genes in the HeLa cell line, which can suggest that GO downregulated cell migration, invasion and adhesion between cells and cells-ECM. In HS-5, *itga3* and *itga5* expression increased, causing up-regulation of cell ability to proliferate and invade, while *itga6* and *itgav* expression decreased, suggesting that adhesion and migration of HS-5 cells were down-regulated. Cancerous and noncancerous cells react differently to GO treatment which can be further studied for potential therapeutic applications.

Keywords: graphene oxide, cervical cancer, integrins, nanoparticles, nanobiotechnology

O-14: Application of bacterial cellulose as a excipient in tablets for oral drug delivery

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Bacterial cellulose (BC) is a biopolymer produced by non-pathogenic bacteria that naturally occur in the environment. Due to its unique properties, such as biocompatibility, high mechanical strength and plasticity, it has been used in various industries, e.g. in medicine or pharmacy. One of the possible uses of BC is its use as an excipient in tablets, which can help in situations where the use of other substances (e.g. sucrose, glucose) is limited in some diseases. The aim of the work was to prepare and characterize BC-based tablets as a excipient for the oral administration of antibiotics. 200 mg tablets based on BC as an excipient and amoxicillin (10, 20 and 30 mg) were prepared using, a standard tablet press. The physicochemical properties of the tablets, such as strength, solubility and absorbency, were assessed. The drug release rate was analyzed in simulated gastric juice (pH 2 and 3.5) and bile salts (pH 5 and 7) during a 4-hour incubation at 36°C, with or without the addition of beads simulating the contents of the digestive system. The amount of released antibiotic was assessed spectrophotometrically. The results were compared with tablets containing wheat starch, lactose, maltose, agar and guar gum as excipients. BC-based tablets showed the best physicochemical properties and the lowest release rate of antibiotics compared to other tested substances. The best solubility for BC-based tablets was observed in the solution with pH between 3.5 and 5. Regardless of the pH of the solution and amount of antibiotic in the tablet, the addition of beads simulating the content of the digestive system increased the solubility of the tablets by an average of two times. Potentially, BC can be used in an oral drug delivery system, especially in cases where a long and stable release of the active substance is required.

Keywords: antibiotic, bacterial cellulose, tablets, oral delivery system

O-15: Droplet microfluidic system for passive selection and enrichment of bacteria producing biosurfactants

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Traditional methods for enrichment and functional screening of microorganisms rely on their growth in a selective liquid medium or on an agar plate, which is followed by tedious characterization. Thus, they can be described as time-consuming, non-quantitative and low-throughput. Some of these limitations are likely to be overcome with droplet microfluidic techniques, which have been recently used to cultivate microorganisms and preserve enriched bacterial taxonomic richness. However, new methods are still needed to select droplets comprising not only growing microorganisms, but also those exhibiting specific properties, such as production of value-added compounds. We describe here a droplet microfluidic screening technique for the functional selection of biosurfactant-producing microorganisms, which are of great interest in the bioremediation and biotechnology industries. Bacterial single cells are first encapsulated into picoliter droplets for clonal cultivation and then passively sorted at high throughput, reaching frequencies up to 250 droplets per second, based on a change of surface tension in individual droplets. Our method expands the droplet-based microbial enrichment with a novel approach and reduces the time and resources needed for selection of surfactant-producing bacteria.

Keywords: biosurfactants, bacteria, microfluidics, surface tension, enrichment

O-16: Seeking for a potent pyocyanin producer using statistical planning

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Pseudomonas aeruginosa is an opportunistic pathogen well-known for producing numerous pigments. The best-studied example is pyocyanin due to its negative role in cystic fibrosis. However, recently it is also recognized as a utile chemical, potentially valuable for energy generation, cancer treatment, or agriculture. For this reason, finding adequate culturing conditions, i.e., temperature, medium composition, or agitation, is no less important than choosing an excellent bacterial producer. Combining both these aspects is crucial to obtain a high yield of the desired product. This work aimed to assess pyocyanin production by different reference strains of *P. aeruginosa* to find the best pyocyanin producer and optimal process conditions. Four tested reference strains (*P. aeruginosa* PAO1, PA14, ATCC 9027 and ATCC 27853) were included in the study. Cells were cultivated in three different media (Tryptic Soya Broth – TSB, Luria-Bertani broth – LB and King's A broth) at three temperatures (27, 32 and 37°C) and three agitation speeds (0, 100 and 200 rpm). After 72 hours of incubation, pyocyanin was extracted from the medium using the chloroform/hydrochloric acid method. Response Surface Methodology (RSM) was used to choose the best pyocyanin producer and optimal process conditions. *P. aeruginosa* PA14 and ATCC 27853 were proven to be the best pyocyanin producers. ATCC 1128 strain was a weak producer in all tested media, while PAO1 did produce pyocyanin, however, in low concentrations and only in richer media. Agitation speed specifically affected the pigment production in tested strains leaving room for process optimization. Both the cultivation conditions and the selection of producer strain should be considered in the pyocyanin production process design.

Keywords: phenazines, *Pseudomonas aeruginosa*, optimization

O-17: Modeling of a putative programmed cell death receptor from *Nostoc punctiforme*

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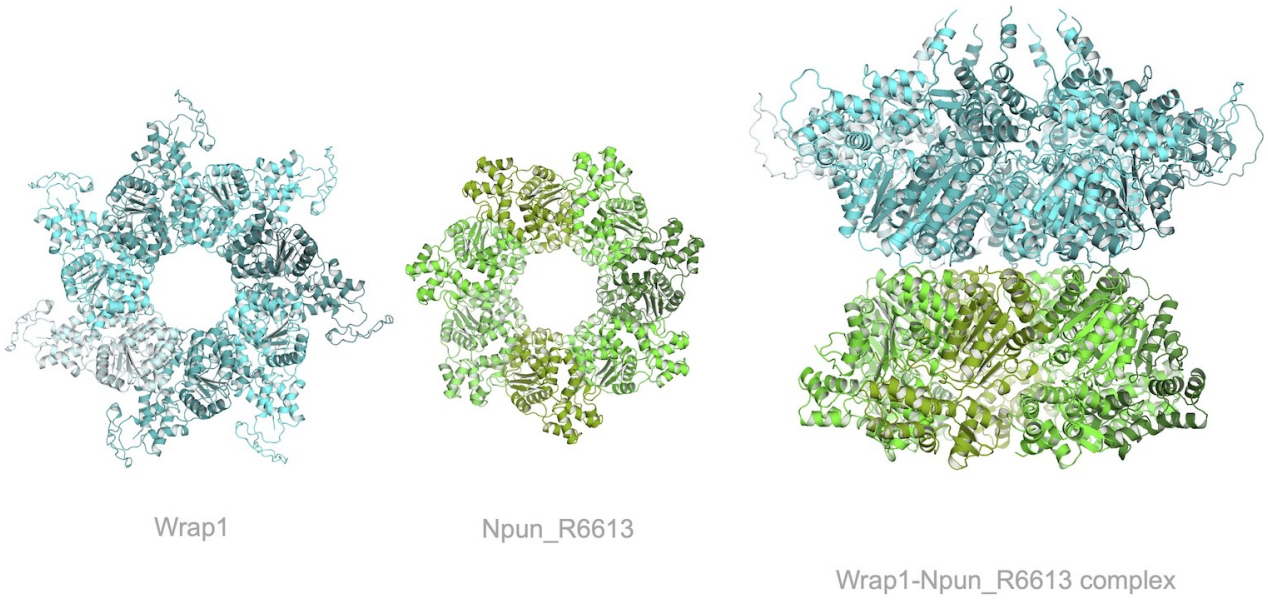
Programmed cell death (PCD) is a mechanism closely associated with multicellularity. It has been reported that well-studied eukaryotic PCD proteins (such as human Apaf-1 or *Caenorhabditis elegans* Ced-4) and not yet fully characterized prokaryotic proteins are homologous. For example, Wrap1 is a transmembrane protein receptor thought to be associated with a putative PCD apparatus in *Nostoc punctiforme* (cyanobacterium). Npun_R6613 is a TIR domain-containing protein encoded in the same operon as Wrap1 and is therefore considered a potential interaction partner. In this study, we computationally modeled the Wrap1-Npun_R6613 complex using AlphaFold2 and Rosetta. To validate the model, we examined the co-evolution between Wrap1 and Npun_R6613 and found that the correlated mutations are consistent with the proposed model. This study extends our knowledge of the prokaryotic mechanisms of PCD and the evolution of the proteins involved. The activity of the Wrap1 complex in inducing PCD will be tested in yeast.

Keywords: programmed cell death, AlphaFold2, Rosetta, protein, bioinformatics



O-18: Kinetic Analysis of IFITs Interactions and the Role of IFITs complexes in IFIT1 Binding to IVT mRNA

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In vitro synthesized mRNA has emerged as a promising drug candidate for various diseases, including cancer, genetic disorders and infectious diseases. For example, mRNA vaccines against COVID-19 represent a significant success in the field of RNA therapeutics. Despite this progress, cellular antiviral mechanisms stimulate high expression of interferon-induced proteins with tetratricopeptide repeats (IFITs) in response to foreign RNA. IFIT1 plays a major role in recognizing and binding to 5' end of nonself-RNA, leading to restriction of foreign RNA translation. Murine IFIT family members of IFIT1, IFIT2 and IFIT3 are known to interact with each other, forming biological hetero-complexes, involved in the regulation of antivirus immune responses. However, the approximate binding affinity constants between these three IFITs and the role of IFITs complexes in the recognition and binding of IFIT1 to different synthesized capped RNA has not been elucidated. Here, we determined biophysical data on the interactions among IFITs and the binding kinetic parameters of IFIT1 to differently capped mRNAs in the presence of IFITs complexes. We analyzed thermal stability of single IFITs and IFITs complexes, demonstrating that IFITs complexes are more thermally stable than single IFITs. MST (Microscale thermophoresis) protein interaction assay proved that IFIT1 shows strong binding affinity to IFIT3 and the highest affinity is observed for IFIT1 to IFIT2/3 complex. Finally, results from BLI (Biolayer interferometry) interaction assay revealed that complex formation among IFITs increase the binding affinity of IFIT1 to mRNA containing cap0 and cap1, providing valuable insights to the understanding of molecular mechanism of mRNA recognition by innate immune system.

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Keywords: IVT mRNA, IFIT1, interaction, mRNA treatment

O-19: Does PD-L1 signaling have an impact in the molecular change in classic Hodgkin lymphoma?

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Classical Hodgkin lymphoma is a type of malignancy originating from abnormal B-lymphocytes characterized by bi and multi-nucleated Hodgkin and Reed-Sternberg (HRS) cells. An important hallmark of HRS cells is overexpression of CD30 and PD-L1 (Programmed cell death protein 1 ligand) on the cell surface. Overexpression of PD-L1 plays an important role in proliferation and metabolism of tumor cells. The modern immunotherapy using anti-PD1/PDL1 antibodies enhances the response of T cell to cancer cells and ultimately blocks the proliferation of HRS cells. For our study we used the certified HRS cell lines: KM-H2, L-1236, L-428. After 3-5 days cells from all harvested lines subjected for Western blot analysis with antibodies: anti PD-L1, EZH2 and SWI/SNF subunits to verify in which line the PD-L1 was overexpressed. The immunoprecipitation was performed on L-1236 HRS cells followed by mass spectrometry analysis. The data obtained from this analysis will be further confirmed by Yeast two hybrid system. Furthermore, we performed chromatin immunoprecipitation (ChIP) on L-1236 cHL line using antibodies: anti SWI/SNF (BAF 155), PRC2 complex (EZH2), PD-L1 as well as methylated and acetylated Histone H3 on CD274 promoter region. We investigated the PD-L1 translocation into cell nuclei and identified its potential nuclear partners in L-1236 cell line. After the mass spectrometry analysis we found PD-L1 may interact in the nucleus with the splicing machinery and thus regulate the RNA posttranscriptional alternative processing. Noteworthy, we subsequently found that the subunits of SWI/SNF (BAF 155), PRC2 complex (EZH2), PD-L1 and methylated and acetylated H3 are located at the same position i.e. (-60) on the promoter region of CD274 gene that encodes PD-L1 protein. Collectively, our findings may provide a clue about the involvement of both EZH2 and SWI/SNF complexes together with PD-L1 itself in regulation of the PD-L1 expression in HRS cells.

Keywords: Classical Hodgkin lymphoma, cells PD-L1, SWI/SNF complex, EZH2

O-20: Mechanistic studies of nonenzymatic self-replication of alternative RNA forms using molecular dynamics

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The RNA molecule (ribonucleic acid) plays a major role in protein synthesis in all organisms (tRNA, rRNA etc.), constitutes ribozymes and is a genetic storage for a group of viruses. Nucleic acids such as RNA have the special ability to direct its own replication process. It is suspected that RNA was important during abiogenesis, because it has catalytic abilities and carries genetic information[1]. However, the way how RNA could have self-replicated without the assistance of enzymes remains uncertain. It was discovered that nucleotides activated with imidazole derivatives may play a major role in non-enzymatic replication on the young Earth, however the A:T pair exhibited the poorest quality of the process[2]. It was also shown that in the presence of 2-aminoimidazole-activated monomers and helper trinucleotides the primer extension was efficient for all 4 canonical nucleotides[3]. In this presentation I will show results of mechanistic studies of the influence of non-canonical nucleotides such as: inosine and 2-thiouridine on the process of non-enzymatic replication of RNA. I had performed classical molecular dynamics simulations of systems containing imidazolium bridged dinucleotides consisting of guanine, inosine and adenine nucleotides; thio-uracil was present in the complementary strand in the system with adenine.

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Keywords: RNA, Self-replication, Molecular Dynamics

O-21: Avidity at its finest: Hsp70 and the art of efficient substrate delivery

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Hsp70 proteins are versatile molecular chaperones, involved in cellular proteostasis and a variety of essential cellular processes by cyclic interaction with substrate proteins. Substrate binding is driven by Hsp70 conformational changes induced by ATP hydrolysis, which is synergistically stimulated by interactions with substrate and a J-domain protein (JDP) co-chaperone. This synergy is not well understood due to the transient nature of both interactions. Here we leverage a well-characterized Hsp70 system, specialized for Fe-S cluster biogenesis. As in many Hsp70 systems, the JDP binds the substrate on its own and then mediates the substrate binding to Hsp70. Based on the established JDP-substrate interaction, we combined molecular docking and molecular dynamics simulations to model the ternary Hsp70-JDP-substrate complex. The model placed the J-domain and substrate in their predicted positions without modulation of the JDP-substrate interaction. To verify the structural model, we isolated the Hsp70-JDP-substrate complex, using an ATPase-deficient Hsp70 variant. We then used hydrogen-deuterium exchange coupled with mass spectrometry to assess the Hsp70 conformation in the ternary complex and to identify the regions stabilized upon complex formation. Our biochemical analyses show that the complex stability depends on previously described residues essential for binary interactions. Furthermore, we identified a novel JDP-Hsp70 binding site that contributes to the complex stability. We conclude that in this system the Hsp70 binding sites of both JDP and substrate are precisely positioned, resulting in super-affinity (avidity), where each interaction site is crucial for efficient formation of the ternary complex. We propose that the precise positioning of substrate on Hsp70, enforced by multiple protein-protein interactions, explains the synergistic ATPase stimulation and efficient substrate binding in many Hsp70 systems.

Keywords: Hsp70, protein-protein interactions, HDX-MS, molecular dynamics

O-22: Autoinhibition in class B JDPs: the key to efficient amyloid fibril disaggregation

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Molecular chaperones of Hsp70 family are critical for cellular proteostasis. They control multiple cellular processes that are essential for cell function, including protein folding and refolding, trafficking, remodeling, disaggregation and degradation. This functional diversity is driven by obligatory partners of Hsp70: J-domain proteins (JDPs). Two main classes of JDPs, class A and class B, are involved in preventing protein disaggregation in eukaryotes. Both JDP classes promote protein disaggregation and support Hsp70 activity by stimulation of ATP hydrolysis. However, only class B JDPs have a unique ability to disassemble amyloid fibrils, the hallmark of neurodegenerative diseases, such as Alzheimer's or Parkinson's disease in humans. Moreover, only class B JDPs are able to bind the C-terminal motif of Hsp70s – EEVD. This difference significantly affects their mode of action, by triggering autoinhibitory mechanism, which allows for multiple Hsp70s to an aggregate and disaggregation. To identify molecular determinants responsible for functional divergence between different JDP classes, we studied the interaction of JDPs with amyloids and amorphous type aggregates using real-time biochemical assays. We show that human class A JDPs efficiently interact with amorphous aggregates and are able to recruit Hsp70 with much faster kinetics than class B. In contrast, human class B JDPs recruit the Hsp70 to amyloid fibrils much more efficiently then class A JDPs. We also proposed a structural model of the autoinhibitory mechanism in class B JDPs, which allows for efficient recruitment of Hsp70s to amyloid fibrils and verify it using site specific mutagenesis. Together, our results elucidate the importance of class B JDPs autoregulation in amyloid fibril disaggregation, a key mechanism in Alzheimer's and Parkinson's diseases.

Keywords: Hsp70, JDP, amyloid fibrils, neurodegenerative diseases

O-23: An ancient tale of two JDPs: The evolutionary journey from protein folding to preventing Alzheimer's disease

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The J-domain protein (JDP) family contains two types of proteins, A and B, that control the correct folding of other proteins in human, yeast and bacterial cells. They function in cellular proteostasis by cooperating with Hsp70 chaperones. Together with Hsp70s, JDPs assist in protein disaggregation and folding by stimulating the ATPase activity of Hsp70 and mediating the substrate binding. Both JDP classes bind amorphous aggregates and fibrillar amyloid aggregates via their C-terminal domain. However, only class B JDPs are able to interact with C-terminal EEVD motif of Hsp70. This interaction allows the dismantling of amyloid fibrils derived from A42 peptide, which are the main cause of Alzheimer's disease in humans. However, the emergence of molecular features of class B JDPs, that allow them to disassemble A42 fibrils, is still unclear. To elucidate the molecular determinants behind the divergence of class B JDPs, I biochemically resurrected the last common ancestor of class A and B JDPs (AncAB) and the ancestor of class B JDPs (AncB). Using biochemical assays, I show that AncAB binds the protein aggregates, similar to modern Class A JDPs. In contrast to Class A JDPs, AncAB is able to bind EEVD motif of Hsp70s, which is a distinctive feature of Class B JDPs. Furthermore, I investigated the interaction of ancestral JDPs with amorphous aggregates and amyloid-type aggregates. My results indicate that AncAB recruits Hsp70s to amorphous aggregates more efficiently than AncB. In contrast, AncB efficiently recruits the Hsp70 system to amyloid fibrils, indicating that JDPs specialised in amyloid disaggregation after the gene duplication. To sum up, my results indicate that functionally divergent class A and B JDPs evolved via gene duplication and subsequent sub-functionalization from ancestral JDP, which shared their biochemical features. This suggests that class B JDPs evolved to prevent the formation of amyloid fibrils and development of neurodegenerative diseases.

Keywords: JDP, Hsp70, ancestral reconstruction, Aβ42, neurodegenerative diseases

O-24: The effect of selected point mutations in spinach violaxanthin de-epoxidase on the course of reaction and enzyme structure

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Violaxanthin de-epoxidase is an enzyme which participates in violaxanthin cycle, a type of xanthophyll cycle, protecting the photosynthetic apparatus in high light conditions. In this study, the effect of P74V, S183T, N187D and D190N mutations in spinach (*Spinacia oleracea* L.) violaxanthin de-epoxidase on reaction kinetics was tested and the structure of the proteins and conservation of substituted residues were analysed bioinformatically. P74V and D190N mutations had the greatest impact on catalysis, while S183T and N187D had a smaller effect. All of the muteins had their three dimensional structures similar to the wild type according to molecular modelling. Interestingly, mutations similar to P74V, N187D and D190N were found in sequences of naturally occurring algal homologues of spinach violaxanthin de-epoxidase and catalytic properties of the studied muteins as well as probable properties of their natural counterparts seem to be matching. **Keywords:** muteins, protein structure, reaction kinetics, violaxanthin de-epoxidase, xanthophyll cycle

O-25: Metabolism of reactive oxygen species in the traps of *Nepenthes x ventrata* during nitric oxide-stimulated digestion

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Nepenthes x ventrata is a carnivorous plant, that forms jug-shaped traps filled with digestive fluid. In the digestive fluid, except for secondary metabolites, digestive enzymes and microorganisms, the presence of reactive oxygen species (ROS) and reactive nitrogen species (RNS) has been demonstrated. Although the occurrence of ROS in the digestive fluid of nepenthes plant traps have been proven, metabolism and localization of ROS in trap tissue is still unknown. The aim of this study was to determine the ROS production in the tissue of the trap and to characterize selected elements of the antioxidant system in the digestive fluid of the pitcher plant (*Nepenthes x ventrata*) during nitric oxide-stimulated digestion. Traps and digestive fluids, collected from unfed and fed traps 24, 48 and 96 h after stimulation were used in the study; sodium nitrate (III) solution was used as the NO_x donor. For the localization of $O_2^{\bullet-}$, the trap tissue was histochemically stained with nitrotetrazolium blue chloride (NBT) and the formed formazan was extracted; to confirm that the NADPH oxidases are responsible for the production of $O_2^{\bullet-}$ in the trap tissue, the effect of diphenylene iodonium chloride (DPI) was determined. The antioxidant system in the digestive fluid was characterised by measuring the activity of peroxidases of class III (POx) and the content of flavonoids. The main results reveals that secretion of $O_2^{\bullet-}$ occurs only in the cells of the secretory glands, DPI effectively inhibited the activity of NADPH oxidases in the secretory glands and NO stimulated digestion increasing the activity of POx and the content of flavonoids in the digestive fluid. The obtained results are fundamental for further research on ROS and their function in the digestive process in the traps of nepenthes plants, it is known that $O_2^{\bullet-}$ is produced by NADPH oxidases in the secretory glands and $O_2^{\bullet-}$ is degraded by the antioxidant system, which is stimulated by NO.

Keywords: carnivorous plant, ROS, NO, NADPH oxidase, POx

O-26: Alterations in ROS metabolisms in the traps of *Nepenthes x ventrata* processing digestion stimulated by nitric oxide

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The ability to use animals as a source of minerals is an adaptation to high-humidity environments with low nutrient availability. Pitcher plants (Nepenthes) belong to carnivorous plants that develop passive pitcher-shaped traps at the end of the main nerve of the leaf blade (Mithöfer, 2011). The traps are filled with digestive fluid in which digestive enzymes, reactive oxygen species (ROS) and reactive nitrogen species (RNS) have been identified (Chia *et al.*, 2004, Wal *et al.*, 2022). In plant physiology ROS, together with RNS, of which nitric oxide (NO) is best characterized, has a regulatory and signalling role. Overproduction of these molecules is toxic to cells, while enzymatic and non-enzymatic antioxidant systems are responsible for maintaining their optimal levels (Hancock and Neill, 2019). The aim of this study was to determine alterations in ROS metabolism in the trap of *Nepenthes x ventrata* processing digestion stimulated by nitric oxide NO_x stimulated digestion. Material (digestion fluid and trap tissue) for the analyses were collected 1, 2 and 4 days after introducing both protein source and NO_x into the pitcher plants' traps. As the control, non-fed traps were used. The formation and removal of ROS by enzymatic and non-enzymatic pathways were analyzed by spectrophotometric and histochemical techniques. The results indicate that NO_x leads to an increase in hydrogen peroxide (H_2O_2)content and total antioxidant capacity, a decrease in superoxide anion ($O_2^{\bullet}$) generation and a decrease in phenolic content in the digestive fluid of pitcher plants. Furthermore, trap tissue staining showed the stimulating effect of NO_x on phenolic content and production of $O_2^{\bullet}$. Oxidoreductive changes in the digestive fluid of pitcher plants may indicate that ROS and RNS play a crucial role in digestion and NO_x may have antioxidant and signalling functions.

Keywords: pitcher plant, digestive fluid, trap tissue, reactive nitrogen species (RNS), reactive oxygen species (ROS)

O-27: Analysis of fatty acid profile in extracts from selected oil rich plant pomace used in *Yarrowia lipolytica* yeast cultures

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Oil rich plant pomace, or there use known as press cake are a side product of the manufacturing process of oils. Due to their chemical composition, namely its protein, fat and polyphenol content, they're a material with high valorisation potential. Currently they are often used as animal feed, but it's considered to be used in microbial cultures. The goal of this study is to attempt to determine the extent to which submerged cultures of *Yarrowia lipolytica* are able to use the lipid fraction of the oil cake. In the study five different variation of oil cake were used: hemp, flax, colza, camelia and safflower. Microorganisms were raised on different substrates, where the additional, or only source of carbon was the lipid fraction of the oil cakes, which were either pulverized or whole. After 6 days of raising the cultures, the composition of fatty acids was analyzed and was compared with fatty acid composition of unprocessed oil cakes. The composition was analyzed using gas chromatography, after previous derivatization into their volatile derivatives (saponification with potassium hydroxide and esterification with methanol).

Keywords: Oil cake, *Yarrowia lipolitica*, Fatty acids, microorganism cultures

P-1: Continuous research work: difficulties and challenges working with primary cell cultures derived from human cancer cells

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Objective: To introduce a continuous research work of difficulties and challenges working in a laboratory with primary cell cultures derived from human epithelial cancer cells. Established cell lines are well analysed in the literature and practice, but primary cell cultures derived from a human cancer tissue could provide us with much more detailed information about particular tumour biology. Unfortunately, surveillance of primary cell cultures induce many challenges for scientists in whole world.

Methods: Small tumor samples from human urinary bladder are obtained to Molecular Oncology department at National Cancer Institute for further steps. Epithelial cancer cells are transferred to a 24-wells plate. On a following days specific growth media is changed depending on wells content physical features and cells viability through microscope. Later on, RNA from primary cell culture, as well as, DNA and RNA from primary cancer samples were extracted for future experiments.

Results: Primary cell cultures are extremely sensitive to various external and internal different factors which could lead to successful or unsuccessful cells development. Some of our samples have not succeed and cells did not grow, thus sample was eliminated. Main reasons of this would be contamination by inner and outer agents or poor cells viability. Luckily, the majority of samples shows great results and we were able to performed RNA extraction.

Conclusions: Thorough and proper work with a high standards of laboratory equipment and researcher hygiene, growth-media quality, non-redundant actions with primary cells cultures are the key to fortunate primary cell growing. When we will be able to grow primary cell cultures which will reflect the primary tumor heterogeneity, we could develop prosperous experiments about individual patient's cancer biology. All information we could possibly gain from these tests could help researchers find new diagnostic and prognostic biomarkers.

Keywords: Primary cell cultures, cancer biology, cancer cells, laboratory work, biomarkers

P-2: Three-dimensional (3D) model of ovarian cancer in Lab-on-a-Chip

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The tumor environment can be described as very complex, complicated and heterogeneous. Taking this complexity into account is definitely important when conducting research on cancer development, diagnosis and therapy. Obtaining a satisfactory *in vitro* model on a macroscale is difficult or even impossible. Designing dedicated Lab-on-a-Chip devices can significantly contribute to solving these problems. The main aim of the research was to create a microfluidic device that simulates the environment of an ovarian tumor. In order to achieve satisfactory results, it was essential to design a structure that would enable the integration of the basic tumor model with vascularization. For this purpose, a microsystem consisting of two parallel, partially connected channels was created. In one of them, a vessel consisting of human umbilical vein endothelial cells (HUVEC) was obtained. The tumor model was created by culturing human ovarian fibroblasts (HOF) and cancer cells (A2780) in co-culture on a membrane located inside the second channel. Various tests were performed to confirm the achievement of the intended goal, including differential staining (to analyze cell viability) and immunostaining (to visualize the complex heterogeneous structure of a cellular model). In conclusion, a microsystem for the simultaneous culture of ovarian cells (non-malignant and cancerous in co-culture) and endothelial cells forming a microvessel was successfully created. After further modifications and improvements, the proposed microsystem can imitate the tumor environment well enough to be successfully used in scientific and diagnostic research.

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Keywords: ovarian tumor, vascularization, tumor environment, Lab-on-a-Chip, microfluidic device

P-3: *Reynoutria japonica* – invasive plant with the potential use in phytoremediation. Study in *in vitro* model

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Anthropogenic pollution caused by urbanization and technological advancements causes huge changes in the natural environment. One of the possible strategies for removing hazardous heavy metals from the soil is phytoremediation, based on the use of plants with broad environmental tolerance. The aim of presented study was to verify whether Japanese knotweed (*Reynoutria japonica*) plants were able to acclimate under stress conditions resulting from the presence of lead. Additionally, an important element of the research was the assessment of *R. japonica*'s ability to accumulate this heavy metal. The research hypothesis assumed that Japanese knotweed, as an invasive plant, has effective defense mechanisms that allow the plant to survive in conditions of heavy metal stress. In the experiment plants were cultivated with the use of *in vitro* cultures with different lead concentrations: 0,0 g/l (control); 0,1 g/l and 0,2 g/l Pb. To estimate biometric parameters of plants under the lead stress growth index and dry weight were analyze. The level of the malondialdehyde (MDA), proline, phenolic compounds (separately for shoots and roots) was measured with the use of spectrophotometric methods. The potential of Japanese knotweed shoots and roots to accumulate lead was examined using flame atomic absorption spectroscopy [FAAS]. The results showed that Japanese knotweed can accumulate lead in roots and shoots. The strategy of acclimatization in lead stress may be related to the protective role of proline and with the mechanism of remodeling the cell wall structure in *R. japonica* plants. This research is the first report about the lead stress tolerance in Japanese knotweed plants giving the strong basis for further research about using the invasive species in phytoremediation.

Keywords: tissue cultures, bioremediation, heavy metals, lead

P-4: Temporary immersion bioreactors and agitated cultures as the useful tool for synthesis biologically active phenolic compounds in *Reynoutria japonica* and its biologically active properties

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Reynoutria japonica is an invasive and environmentally dangerous species, but at the same time can synthesize many polyphenols. This group of plant secondary metabolites has a wide biological activity and potential application in the medical field. In natural habitat *R. japonica* occur in contaminated area and may accumulate harmful elements in the tissue, so obtaining these plants from the environment is limited. Therefore, in this study for the first time *R. japonica* plants were cultivated in *in vitro* conditions. To increase accumulation of biomass and synthesis of polyphenols temporary immersion bioreactors Plantform™ (TIB) and agitated cultures on rotary shakers (AC) were used for the cultivation. The control in experiment were tissue cultures of *R. japonica* grown in traditional agar-solidified medium (A-SM). The main hypothesis of the study was: that tissue cultures of *R. japonica* may be an efficient source of biomass rich in phenolic compounds, while the use of TIB and/or AC for plants cultivation may increase biomass accumulation and enhance synthesis of secondary metabolites. Moreover, we hypothesized that extracts obtained from plants cultivated in TIB and/or AC will be characterized by strong antioxidant and bactericidal properties. Grown plants from A-SM, AC and TIB were analyzed for the growth index and dry weight estimation. Plant tissue were extracted in 80% methanol (separate for shoots and roots) for the analysis of total phenolic content and antioxidant activity using spectrophotometric methods with Folin&Ciocalteu's reagent and DPPH radical, respectively. Moreover, analysis of selected polyphenols accumulation was performed by DAD-HPLC. Also, antibacterial properties of examined extracts were expressed as the minimal bactericidal concentrations of tissue for *Staphylococcus aureus* ATCC 25923. Results have shown that TIB and AC are more effective platforms for synthesis of biologically active phenolic compounds in *R. japonica* than A-SM.

Keywords: polyphenols, bioreactors, *in vitro* cultures

P-5: The nanostructure of the polymeric solution determines the delivery of macromolecules into cells via osmotic shock

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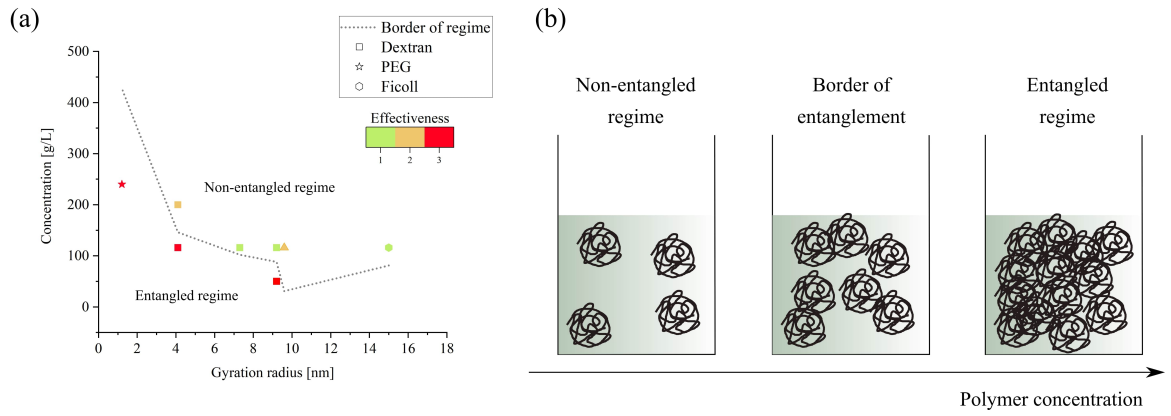
Crossing the cell membrane is a limiting step in most biochemical or biomedical research. There are many techniques that allow the introduction of a desired molecule into cells. One of them is the application of osmotic shock. Its principle is based on water flow from a hypotonic solution (with a lower osmotic pressure) to the hypertonic one (with a higher osmotic pressure) through a cell membrane. The hypertonic medium includes a polymer that builds osmotic pressure. We analyzed how the concentration and the size of a polymer in the hypertonic solution impact the effectiveness of intracellular delivery via osmotic shock. As tested osmotic polymers, we chose different dextrans, PEGs and Ficoll. Our studies clearly showed that the entangled regime is a crucial parameter determining the effectiveness of cargo delivery using osmotic shock. As a result of our research, we have developed an optimized polymeric formulation called Cell-IN that enhances the cellular uptake of a desired molecule. The effectiveness of the Cell-IN has been confirmed for a wide range of cargos (dyes, polymers, proteins, nucleic acids, nanoparticles) and cell types (normal, cancer, epithelial, mesenchymal cells).

Keywords: cellular delivery, osmotic shock, entangled regime, pinocytosis, nanorheology



P-6: Study of ATP bioluminescence metres in assessing decontamination of biotechnological laboratories

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The maintain safety standards in biotechnology laboratories is required during every scientific research. Avoiding contamination by microorganism from air, water and surfaces is crucial for the staff safety and their workplace. A varied microbiology methods are used in order to detect a potential hazards. One of these methods is luminometric determination of adenosine 5'-triphosphate (ATP). It is commonly used for assessing environmental cleanliness and microbiological pollution of surfaces. The main goal of conducted research was determination of luminometric methods for assessing microbial air quality at biosafety level 1 (BSL-1) biotechnological laboratory. In addition, the conventional technique of open plates (Koch sedimentation method) was used. Studies were conducted over four weeks, at five research sites, located in five different places at biotechnological laboratory. To measures the strength between the different variables and their relationships Pearson correlation coefficient was calculated.

Based on the results obtained at the BSl-1 biotechnological laboratory (Department of Chemical and Process Engineering, WUT) the enormous number of microorganisms were detected. The air quality does not meet the standards for drug laboratories. In the air three times more bacteria than fungi were detected. There is direct correlation between ATP concentration (RLU) measured by luminometric method and the number of microorganisms [CFU/m^3] in the air estimated based on Koch sedimentation method. This study examined the feasibility of using adenosine triphosphate (ATP) bioluminescence at biotechnology laboratory. It can be used as a rapid and effective tool for monitoring surface cleanliness in real time.

Keywords: luminescence, ATP, microbiological air quality, biotechnology laboratory

P-7: Purification of air from mixture of volatile organic compounds by adsorption method

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When considering the topic of air pollution, it is mainly CO_2 and its potential impact on the environment that is discussed. Less is said about volatile organic compounds, which are emitted by many industries or even plants. Moreover, people are exposed to the harmful effects of these compounds at home, at work, or in the car. Hence the importance of making the public aware of the harmfulness of these compounds. As scientists, we should strive to find effective and economical methods of air purification that will enable us to breathe cleaner air. Our task is to deepen and supplement scientific knowledge, which will contribute to developing industrial air purification processes. Therefore, the aim of the presented work is to describe the adsorption process of a mixture of VOC vapors on activated carbon and to determine the effect of modifying the adsorbent with malic acid on the process. A vapour mixture was adsorbed: acetone, ethyl acetate, toluene and n-butyl acetate. The properties of the adsorbents were investigated by Boehm titration and the iodine number, ash content and elemental composition were determined. A mixture of air and VOC vapor was passed onto the adsorbent bed. The outlet concentrations of the adsorbates were continuously monitored by gas chromatography with a flame ionization detector. The results obtained allowed the calculation of the adsorption volume of individual adsorbates and the migration velocity of the sorption front. The Yoon-Nelson kinetic model was satisfactorily fitted to the experimental data. The phenomenon of competitive adsorption was observed in the studied system. The volatility of the adsorbates was found to have a crucial influence on the adsorption of this mixture. The chemical compound with the lowest vapor pressure adsorbs to the greatest extent. Modification of the adsorbent with malic acid increased the carbon content. In addition, it decreased the ash content, which increased the total adsorption capacity by 22%.

Keywords: multicomponent adsorption, air purification, modified activated carbon

P-8: Antibiotic resistance of *Salmonella* sp. bacteria isolated from sewage sludge from the Silesina, Opole and Lower Silesian voivodenships

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Sewage sludge and its use as biomonitoring are a direct indicator of the health status of a region's population. One of the many indicators may be the presence of *Salmonella* spp. in the sediment, indicating infection, because it is excreted with the faeces and after their detection and isolation, they can not only be identified, but also their resistance status can be checked. It has long been known that wastewater contains antibiotic residues and resistance genes that cause selection pressure in the bacterial environment. This can lead to complications in the treatment of bacterial infections and also lead to the formation of new resistance cassettes. Bacteria of the genus *Salmonella* cause numerous infections in humans and livestock. Most often it occurs in the form of acute food poisoning. Its reservoir can be both animals (domestic and breeding) and people. In acute disease states, it is recommended to use antibiotic therapy after early identification of the strain from the faeces, but with a milder clinical picture, antibiotic treatment is not recommended due to the increasing cases of infections with *Salmonella* strains resistant to ampicillin, trimethoprim or chloramphenicol. The poster presents the current results of research on the resistance of *Salmonella* bacteria isolated from sewage sludge in selected voivodeships in Poland. Of the 244 samples taken, 54 positive samples were obtained, tested by real-time PCR and confirmed by the horizontal method in accordance with the methodology included in the ISO 6579-1: 2017 standard. Samples were collected and checked in the period from June 2022 to January 2023, they were tested for resistance status using 6 different antibiotics on paper discs (ampicillin 25mcg/disc, tetracycline 30mcg/disc, streptomycin 25mcg/disc, ciprofloxacin 5mcg/disc, amoxycillin/clavulan acid 20/10mcg/disc and meropenem 10mcg/disc).

Keywords: *Salmonella*, antibiotic resistance, biomonitoring, antibiotics, sewage sludge

P-9: Exploring interactions between short RNA ended with synthetic cap analogs and an innate immune response protein IFIT1, using bio-layer interferometry technique

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mRNA-based technology has been developing for decades, especially in the medical context. As early as the 1990s, the first experiments on protein complementation through the delivery of mRNA template were undertaken. At that time, the first attempts were also made to use an antigen encoded in mRNA as a new form of vaccine. A breakthrough moment in the history of mRNA technology came during the COVID-19 pandemic, when the mRNA-based approach was used to create two effective vaccines in a relatively short period of time, demonstrating its high potential. Key aspects of therapeutic mRNA, besides its delivery to the cell, are its viability and translation efficiency. One of the important structural elements of eukaryotic mRNA affecting these parameters is the cap structure at its 5' end. To date, most studies have focused on the interaction of cap and its analogs with the eIF4E protein, which initiates translation. The aim of this study was to investigate the interactions of new cap analogs with the human IFIT1, a protein that is part of the innate immune system at the intracellular level. IFIT1 binds to cap structures of mRNAs that differ from the host cap in their methylation pattern and thus blocks eIF4E access to such particle, so that translation cannot be initiated. This action is part of the innate immune response against simple RNA viruses. The data presented here were obtained using bio-layer interferometry in visible white light. This is a label-free technique that does not require any special absorption or fluorescence properties of the particles under study. Instead, it relies on measuring changes in the optical path length in the layer of associated particles.

Keywords: IFIT1, mRNA, cap, innate immune system, bio-layer interferometry

P-10: Changes in morphology and physiology of streptomycetes after contact with nanomaterials

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Streptomycetes are microorganisms commonly found in soil and aquatic environments. The adaptivity of these bacteria to different environments give them high biotechnological potential. They produce many clinically-used antibiotics, e.g., streptomycin, cephalosporin, tetracycline, neomycin and daptomycin. Moreover, these bacteria can also produce numerous biologically active pigments and industrially utile enzymes (e.g., transglutaminase). Recently, it was suggested that nanomaterials can alter bacterial viability and secondary metabolism but little is known about their effects on streptomycetes. Therefore, this study aims to investigate the changes in the morphology and physiology of *Streptomyces* strains contacted with nanomaterials. The study used *Streptomyces griseus* ATCC 10137 (as a reference) and four wild-type *Stretomyces* spp. The effects of nanomaterials (carbon nanotubes, copper functionalised carbon nanotubes, graphene oxide, graphene oxide with cobalt, mesoporous silica nanospheres with titanium dioxide and mesoporous silica nanotubes with titanium dioxide) on colony and cell morphology, biomass production and secondary metabolism were investigated. Deviations in *Streptomyces* colony morphology depended on the used nanomaterial. Furthermore, biomass have shown great affinity to nanomaterials. The toxicity of nanostructures containing copper particles could be visualized in cytograms generated by flow cytometry. Functionalised nanomaterials, i.e., those containing copper, cobalt, or titanium dioxide nanoparticles, resulted in a statistically significant increase in dye production. These observations could be further developed and potentially applied to biotechnological processes.

Keywords: *Streptomyces*, nanomaterials

P-11: Development of the method to lead immobilization in hydroponic cultivation of plants using Ureolytic bacteria *Ochrobactrum* POC9

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Lead accumulation in agricultural soil is harmful to human health and the environment, thus methods to prevent or reduce lead uptake by plants are of great interest. Microbial-induced carbonate precipitation (MICP) is a method for immobilizing heavy metals in contaminated soils using ureolytic bacteria able to NH_4^+ and CO_3^{2-} production by hydrolyzing urea. Carbonate ions react with heavy metals and form less mobile and less toxic precipitates. Moreover, the NH_4^+ produced in the process can serve as a source of nitrogen for plants. The aim of our studies was the development a method for effective immobilization of lead potentially available for hydroponically grown plants, with the use of the ureolytic bacterium *Ochrobactrum* POC9. This strain was grown on medium with urea for 3 days and then the supernatant was used to supplement Knopp medium (used for plants cultivation) with various amounts of lead. Various parameters of the mixture of Knopp medium with POC9 metabolites were verified to optimize of MICP method efficiency. For this purpose, various concentrations of lead (5, 12.5, 25 and 50 mg/L), initial pH values of Knopp medium (5.5, 6, 6.8, 7.5, 8.1 and 8.9) and amounts of added metabolites (1%, 2%, 5%, 10% v/v) were tested. We showed that an initial pH of 7.5, 12.5 mg/L of Pb and a 2% v/v of POC9 metabolites resulted in the highest precipitation ratio, the most stable precipitate and minimal impact on other elements' availability. Further experiments will involve hydroponically grown plants to evaluate lead uptake and the impact of metabolites on their growth. The precipitation of lead in hydroponic conditions is the initial step towards an effective approach for bioremediation and immobilizing this element in agricultural soil conditions, as well as the promotion of crop growth.

Funding: Project No. $LIDER/13/0051/L-11/19/NCBR/2020$ funded by the National Center for Research and Development (Poland)

Keywords: Carbonates, precipitation, ammonia, agriculture, plant uptake

P-12: Analysis of the role of extracellular vesicles in the transmission of ferritin from human macrophages to cancer cells

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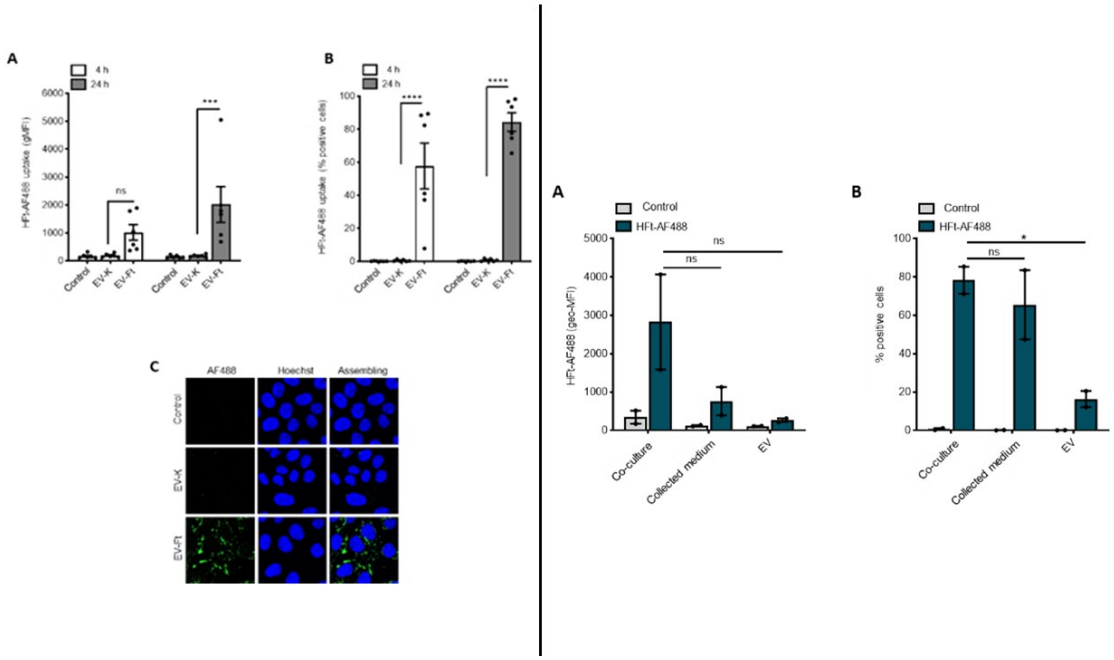
Breast cancer is one of the most common cancers in the world. Conventional treatments have low efficacy or cause too high side effects, so there has been a need to seek alternative anti-cancer therapies. One of the potential methods is a cell-based therapy which makes use of macrophages, as carriers that precisely deliver ferritin molecules in a complex with an anti-cancer drug to cancer cells. However, in order for such a therapy to be used in the treatment of cancer patients, it is necessary to precisely characterize the mechanism of ferritin transfer to cancer cells. One possible option is the transport of this protein from macrophages to tumor cells via secreted extracellular vesicles (EV). The aim of this study is to determine the role of EV secreted by macrophages in delivering ferritin to MDA-MB-231 cells. The conducted research uses the Western blot method, flow cytometry and confocal microscopy. On the basis of the obtained results, it was found that the transport of HFt with the use of EV secreted by macrophages is not the main mechanism of the delivery of this protein to cancer cells.

Keywords: breast cancer, macrophages, H-ferritin, extracellular vesicles



P-13: Analysis of the survivality of *Listeria monocytogenes* under bacteriophage pressure in digestate, produced in biogas plants

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Bacteriophages (phages) are currently in the spotlight due to the emergence of multidrug-resistant bacteria. Because of specificity and ecological safety, phages are a promising alternative in the treatment of bacterial infections, this is due to the fact that they are their natural killers and show very high efficiency in the regulation of bacterial populations. The overwhelming majority of papers focused on bacteriophages based on determining their application potential for biomedical applications. The potential use of phages in sanitation processes in a form dedicated to biogas production processes has not been described so far. Post-fermentation waste (digestate), especially from agricultural biogas plants, undergoing subsequent transformations, may contain dangerous pathogens like *S. Typhimurium*, *Y. enterocolitica*, or *L. monocytogenes*, which may contaminate the natural environment.

In this research, we determined the effect of the addition of phages on the formation of the *L. monocytogenes* population in the digestate from an agricultural biogas plant. To determine the initial number of this bacterium in the post-fermentation waste, a microbiological analysis was performed using the chromogenic media and then the lytic activity of phages was checked on the isolated strains using a modified spot-test method. The next step was to add bacterial viruses to the samples and incubate them for 6 weeks to determine the changes. Each week the number of bacteria and specific bacteriophages was checked and calculated. The obtained results show the possibility of using phage preparations in a specific environment – digestate. A statistically significant reduction in the number of bacterial cells was observed, both for reference and naturally occurring strains. The increase in the number of virions was observed each week, which proves both their stability in a specific environment and the fact that they can carry out an effective replication cycle.

Acknowledgements: This work was funded from grant $LIDER0069/L-12/2020$

Keywords: Bacteriophage, Microbiology, Biogas

P-14: Nitrogen cycling model in Arctic ornithogenic soil (Hornsund, Svalbard)

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Arctic is changing rapidly due to global warming. Thawing of permafrost, the concomitant release of organic matter and the continuous increase in temperature, accelerate the metabolism of microorganisms, including denitrifies and nitrifiers which contribute to emissions of a highly potent greenhouse gas - N_2O. Although polar regions are considered oligotrophic ecosystems, emissions from hot spots in the Arctic (including cryoturbated soils and peat circles) have been recognised as one of the highest from natural sources. Recently, ornithogenic soils were also found to harbour this potential due to the significant input of N caused by the activity of bird colonies. In this study we investigated biogeochemical N cycle in different soil habitats in vicinity of the Polish Polar Station Hornsund, to identify areas with high potential to release GHG. In examined sampling sites, high levels of organic and inorganic nitrogen compounds were identified in ornithogenic soil, including amino acids and ornithine cycle substrates, NH_4^+, NO_3^- and NO_2^-. Moreover, total N and C content were the highest there and C:N ratio the lowest (3,65), which are conditions promoting N_2O emissions, further confirmed using GC-MS/SIM method. This habitat also exhibited the highest bacterial diversity and abundance of bacteria involved in N turnover, most importantly nitrifiers (*Nitrospira*) – reducing NH_4^+ and being able to produce N_2O under microaerophilic conditions and denitrifiers (e.g., Bradyrhizobiacaea family members) – reducing products of nitrification to N_2O or N_2 in anoxic environment. Also, N-fixers (*Cyanobacteria* and *Rhizobiales*) and bacteria involved in the annamox process (*Gemmataceae* family, phylum *Planctomycetes*) were identified. All these features collectively hint at soils of ornithogenic origin as being hot spots for the highly potent N_2O gas emission. Based on those results we constructed a model of N cycling in ornithogenic soils, which can be useful in future predictions.

Keywords: Arctic, soil, bacteria, nitrogen, GHG

P-15: Index patient in an alimentary tick-borne encephalitis (TBE) outbreak

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Introduction: Tick-borne Encephalitis virus (TBEV) can lead to Tick-borne Encephalitis (TBE), a serious neurological infection. TBEV is mainly transmitted through tick bites but in 1% of cases, it is transmitted by the consumption of unpasteurized dairy products from domesticated ruminants. Alimentary TBE infections have a shorter incubation period, less severe course and occurs sporadically and in small outbreaks.

Case presentation: A 69-year-old female patient was admitted to our department in the context of fever up to 39°C, severe headache, hand tremor, nausea and vomiting for the past 24 hours. Two weeks earlier she consumed unpasteurized goat milk with six members of her family, in which they all developed flu-like symptoms a few days afterward. Upon admission, the patient was conscious but slightly slowed down and confused. CSF analysis revealed lymphocytic pleocytosis (55 cells/µl) and high protein levels (70.1 mg/dL). PCR of CSF with BioFire FilmArray Meningitis/Encephalitis panel was negative. Suspicion of alimentary TBE was raised due to the patient's history and clinical profile. ELISA TBEV-IgM was positive in both serum (4.20 g/L) and CSF (4.45 g/L) confirming the diagnosis of TBE. She was treated with intravenous Dexamethasone and started rehabilitation therapy. Over the course of treatment, her symptoms improved gradually and ten days after admission the patient was no longer complaining of any symptoms and was rehabilitated without neurological complications.

Conclusion: This outbreak highlights the importance of proper pasteurization of dairy products as food safety measures cannot be overemphasized in preventing such illnesses. In addition, it highlights the effectiveness of early use of steroids in reducing neurological complications in certain cases of TBE.

Keywords: Tick-borne Encephalitis, Alimentary TBE, Milk-borne infections, TBE outbreak

P-16: Evaluation of the biocidal activity of nanoparticles and their synergistic effects in the control of protothecal bovine mastitis

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One of the most common diseases of dairy cattle is mastitis. The occurrence of mastitis has been associated mainly with bacteria and fungi, however, in recent years new cases caused by achlorophyllous algae of the genus *Prototheca* have been reported. Currently, no effective method has been developed to combat the algae that cause protothecal mastitis due to their resistance to antibiotics. The aim of this study was to determine the effect of gold (AuNPs), silver (AgNPs), copper (CuNPs) nanoparticles (NPs) and their complexes, on the viability of *Prototheca* spp. Milk from the cattle diagnosed with mastitis was seeded onto a Oxoid Sabouraud Dextrose agar (Thermo Scientific, United Kingdom) and incubated for 24 hours at 37°C. Material from the cultivated colonies was diagnosed using transmission electron microscopy (TEM) images. The suspension of isolate was pipetted into a 96-well plate at a volume of 50 µl per well. The experiment used commercially available AuNPs, AgNPs, CuNPs (Nanotech, Warsaw, Poland). Then 50 µl each of nanoparticle hydrocolloids were added so that the final NPs concentrations were: 1.56, 3.125, 6.25 ppm. Incubation was carried out for 24 hours at 37°C. After 24 hours, 10 µl of Presto Blue reagent (Invitrogen, Waltham, MA, USA) was added to each well and the absorbance was read spectrophotometrically. The conducted study confirmed that AuNPs, AgNPs and CuNPs have biocidal activity against *Prototheca* sp., however, their effects varied according to their concentrations. The highest concentration tested for AuNPs was 6.25 ppm and this reduced the viability of *Prototheca* spp. by 91.5%. All nanocomplexes reduced pathogen viability which confirms their synergistic effect. The AgCuNP complex was the most active, at concentration of 6.25 ppm, which decreased pathogen viability to 1.23%. The study suggests that AgCuNPs may provide an alternative to antibiotics in the treatment of protothecal mastitis, however further *in vivo* experiments are required.

Keywords: mastitis, nanoparticles, nanocomposites, dairy cows

P-17: Impact of copper nanoparticles and cisplatin on HeLa cervical cancer cells line and U251 glioblastoma cells line viability

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The fight against cancer is one of the most serious challenges of modern medicine. The development of science makes it possible to find new solutions and improve the existing ones. Cisplatin, one of the most popular chemotherapeutic, combined with nanoparticles can act more specifically while reducing the number of side effects. The purpose of this study is to determine the effects of copper and cisplatin nanoparticles and their biocomplexes on the viability of cervical cancer cells and stage glioblastoma. Single treatment of tumor cells with copper nanoparticles did not affect the effect of cisplatin, compared to samples treated with the chemotherapeutic alone. Treating glioblastoma cell with copper nanoparticles three times significantly increased its sensitivity to cisplatin – mortality increased by 90% compared to controls and by 38% compared to the group treated with cisplatin alone.

Keywords: copper nanoparticles, cisplatin, HeLa, U251, glioblastoma, cervical cancer

P-18: Lipolytic and proteolytic activity of antarctic bacteria

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Microorganisms living in many diverse locations produce enzymes degrading lipids and proteins. Study of unique bacterial strains obtained from isolated areas can lead to the discovery of new biological catalysts, which could find use in industry or research. [1], [2]. The aim of the study was to determine if selected strains of antarctic bacteria produce substances degrading proteins or lipids. Their activity was tested with media containing skimmed milk, gelatine or polysorbate. Six different strains of bacteria have been selected for the experiment. In order to test for proteolytic activity, they were grown on six types of solid media containing gelatine or skimmed milk in varying concentrations. Visible clear halos around some of the colonies indicated protein degradation. To check for lipase production, the strains were inoculated on two different nutrient media with added Tween 20 or Tween 80. A white precipitate formed around bacteria revealing lipolytic activity. The results show that all strains demonstrate lipolytic activity. Most of them exhibit proteolytic activity as well. The intensity of the effect varied between particular strains and its visibility depended on media composition. Further analysis will enable identification and separation of substances produced by the bacteria, which may lead to isolating previously unknown biological catalysts.

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Keywords: Antarctica, antarctic bacteria, lipases, proteases, bacterial enzymes

P-19: When glial cells encounter hypoxia-ischemia – the involvement of autophagy

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Glia constitute half of the cells building the nervous tissue of the brain. They play an essential role during the formation and proper functioning of the central nervous system. Glia are also involved in pathophysiology as well as neuroregenation in various pathological brain conditions. Such condition, that affects brain in its developing state, is neonatal hypoxia – ischemia (HI). Transient undersupply of oxygen and nutrients to the nervous tissue can cause the long-term effects. HI can lead to brain injury and inhibited brain development, especially the white matter, which results in cognitive and motor disorders. There are several mechanisms, that may be involved in the progression of tissue damage, like oxidative stress or inflammation. However it is unknown whether dysregulated autophagy process, proven to cause neural cell death in this condition, can play an important role in the response of glial cells to the altered tissue homeostasis. In the study, we used the three glial fractions: microglia, astrocytes and oligodendrocytes, isolated from neonatal rat brains and exposed them to the short deprivation of oxygen and nutrients in the *in vitro* culture. We analyzed the effect of the procedure on the expression of markers of autophagy – LC3B and p62, lysosomes and cell viability. We observed a decreased expression of p62 in astrocytes 6h and 24h after OGD and decreased expression of p62 in oligodendrocytes and microglia 6h after the injury. It may indicate the enhanced autophagy process, which was verified with immunolabelling of autophagosomes and autophagolysosomes. PrestoBlue assay revealed decreased viability of cells shortly after the injury. These results suggest that targeting autophagy in glial cells after HI could be an interesting new therapeutic strategy to modulate glial cell response after neonatal asphyxia.

Keywords: Autophagy, hypoxia-ischemia, glial cells

P-20: A yeast-based system for studying interactions between human ACE2 and the Spike protein of SARS-CoV-2 coronavirus

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The COVID-19 pandemic has prompted investigations into the interactions between SARS-CoV-2 proteins and host cells. Human angiotensin-converting enzyme 2 (hACE2) has received considerable attention due to its crucial role as a receptor that allows SARS-CoV-2 to infect human cells. This study aims to establish a yeast-based system using *Saccharomyces cerevisiae* to investigate the interaction between hACE2 and the receptor-binding domain of the SARS-CoV-2 Spike protein (Spike-RBD) from different virus variants. To create this system, we used modified Yeast Surface Display (YSD) technology in which hACE2 is fused to the yeast surface protein a-agglutinin, allowing exposure of the recombinant protein outside the cell. Our yeast cells expressing hACE2 also show red fluorescence and can be incubated with the green fluorescent EGFP-Spike-RBD fusion protein. The interaction between hACE2 and Spike-RBD proteins can be evaluated by changes in fluorescence intensity at different emission wavelengths. Such changes may result from alterations in hACE2 and Spike-RBD interactions. Our system could provide a better understanding of the virus-host relationship, leading to more informed research and drug development, particularly with regard to interactions between hACE2 and the SARS-CoV-2 Spike protein.

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Keywords: *Saccharomyces cerevisiae*, YSD, hACE2, Spike

P-21: Antioxidant properties of *Aralia racemosa* and *Aralia spinosa* (*Araliaceae*) methanolic extracts

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Aralia racemosa and *Aralia spinosa* are plants belonging to the family *Araliaceae*. Both plants were used in folk medicine to treat various diseases (including skin diseases). Little research has been done on the medicinal properties of *A. spinosa* and *A. racemosa*, however the anti-oxidant, antidiabetic, anti-inflammatory and analgesic properties of *A. racemosa* extracts and antimicrobial properties of *A. spinosa* extracts have been reported. The aim of the study was to compare anti-oxidant potential of callus tissue of both species. The callus cultures were established from leaves of *in vitro* growing plantlets of each species. The callus was cultivated on solid MS medium supplemented with 0.5 or 1 mg/L of 2,4-D for 4 weeks. Next, the callus was collected and lyophilized and methanolic extracts were prepared. Total phenolic compound content (TPC) and total flavonoid content (TFC) were determined, as well antioxidant activity using DPPH radical scavenging method. The effect of culture variants on TPC and antioxidant activity varied depending on *Aralia* species and 2,4-D concentration (Fig. 1B,C) and were highly corelated – Pearson correlation coefficient amounted to R^2=0.95. Whereas TFC was not affected by culture conditions (Fig. 1A).

Keywords: *Aralia racemosa*, *Aralia spinosa*, callus culture, antioxidant properties, 2,4-D

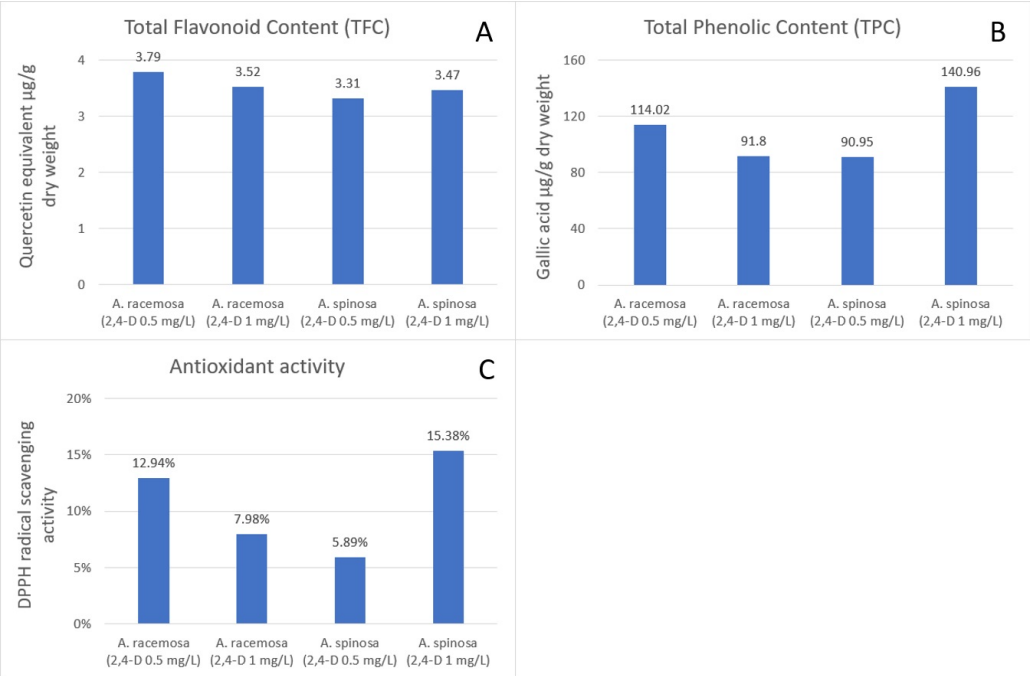

Fig. 1. The effect of culture variant on biological properties of Aralia's callus tissues

P-22: The effect of the lack of dystrophin and its individual isoforms on the proliferation and differentiation of the SH-SY5Y human neuroblastoma cell line

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Duchenne muscular dystrophy (DMD) is an incurable multi-systemic chromosome X-linked disease that affects mostly boys. Mutations in the dystrophin (DMD) gene lead to loss of dystrophin protein. The DMD gene contains several tissue-specific promoters, thus forming different isoforms with different lengths and sites of expression. Results of recent studies show that the Dp140 isoform predominates in the foetal brain and disappears during adolescence. Furthermore, Dp140, along with the Dp71 isoform, was indicated to be involved in the dendrite development and neuronal differentiation. The purpose of the study was to: 1) evaluate expression of the DMD gene in proliferating cells and at various stages of neuronal differentiation, 2) determine whether and how lack of dystrophin affects neuronal differentiation and 3) assess the role of distinct dystrophin isoforms in neuronal cells. To address these aims, we used an SH-SY5Y human neuroblastoma cell line with the potential to differentiate into neurons. RTqPCR was employed to evaluate the presence of dystrophin isoforms during neuronal differentiation and the CRISPR-Cas9 technique was used to generate dystrophin-deficient SH-SY5Y cell line. Furthermore, expression vectors carrying splicing variants of distal dystrophin isoforms were generated to verify their localisation and function in neural cells. The current data show that during neuronal differentiation the full-length dystrophin is synthetized at relatively constant but decreasing level. As expected, Dp140 isoform was present at the early stages of differentiation. Additionally, we succeeded in generation of dystrophin-deficient SH-SY5Y cell line. The current experiments focus on comparison of the proliferation rates and differentiation of control and dystrophin-deficient SH-SY5Y cell line as well as verifying the roles of splicing variants of distinct dystrophin isoforms following delivery of the expression vectors to the dystrophin-deficient SH-SY5Y cell line.

Keywords: Duchenne muscular dystrophy, dystrophin Dp140, SH-SY5Y cell line, CRISPR-Cas9

P-23: Identification of novel inhibitors of the main protease of SARS-CoV-2 using a yeast-based system

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The SARS-CoV-2 pandemic has caused the deaths of millions of people and left its mark across the globe. It also reminded us that the next pandemic could occur at any time. Despite the rapid introduction of SARS-CoV-2 vaccination that has significantly improved patient outcomes and reduced the spread of the disease, many parts of the world still lack access to the vaccine. The search for effective drugs against the virus is therefore crucial. We decided to develop a yeast-based system to screen for inhibitors of the SARS-Cov-2 main protease (Mpro). We used the CRISPR system for the elimination of multidrug resistance and for the inducible expression of active and highly cytotoxic Mpro. The resulting strain was then used for a screening test on solid medium using almost 2,000 drugs. As a result of the tests, a group of promising Mpro inhibitor candidates was selected. One compound shows potent Mpro inhibitory activity and its mechanism of action appears to be different from all currently used drugs against COVID-19. This suggests the possibility of a new treatment approach for the disease. In conclusion, the identification of potential drugs for the treatment of COVID-19 using the yeast-based system developed for the screening of Mpro inhibitors is promising and could serve as a starting point for further research into the development of drugs that are effective against multiple variants of the virus.

This work was supported by the research grant of Adam Mickiewicz University, Poznan, Grant Number 6/2020"$ResearchonCOVID-19$".

Keywords: SARS-CoV-2, *Saccharomyces cerevisiae*, Mpro, main protease, drug screening

P-24: Evaluation of the effectiveness of bacteriophages in the *Staphylococcus aureus* and *Pseudomonas aeruginosa* bi-species biofilm

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Due to increasing difficulties in the treatment of antibiotic-resistant bacterial infections, the research for alternatives and effective treatment methods is gaining popularity. In recent years, therapy using phages, which are viruses specifically infecting bacteria, being intensively studied. One of the most common etiological factors of infections are antibiotic-resistant bacteria of the species *Staphylococcus aureus* and *Pseudomonas aeruginosa*. This work aimed to analyze the phage infection in the system species biofilm of *P. aeruginosa* and *S. aureus* with the determination of population changes in biofilm components. To conduct the research, biofilm cultivation was started using starter cultures of *P. aeruginosa* and *S. aureus* after overnight incubation. The study was conducted in systems enabling the determination of interactions between all tested factors: *P. aeruginosa* and *S. aureus* bacteria and infecting them with phages. To characterize the biofilm, staining methods were used using resazurin and crystal violet. To determine the impact of phage pressure on changes in the structure of the population, the analysis of components was conducted using breeding methods on dedicated substrates. The results show high bacterial biofilm reduction activity produced by *P. aeruginosa* and *S. aureus*. In addition, in the studied model of a two-species biofilm, niche-taking over by *P. aeruginosa* was observed, when wasn't subjected to phage pressure. However, in the case of adding to the culture bacteriophage infecting *P. aeruginosa*, the composition of the population changes a shift in favor of the number of *S. aureus* occurring. Using phage therapy consisting of one phage, in a model of unknown full infectious etiology, may lead to taking over by another bacterium. Moreover, bacteria that become resistant to phage infection may show other phenotypic properties.

Funding: National Science Centre, Poland PRELUDIUM 19, Project No. 2020/37/*N*/*NZ*9/02947.

Keywords: phages biofilm antibiotic-resistant, infection

P-25: Biological activity of secondary metabolites extracted from plants belonging to genus *Helleborus* spp.

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Background: *Pseudomonas aeruginosa* and *Staphylococcus aureus* are microorganisms belonging to the ESCAPE group - of particularly troublesome pathogens possessing the ability to develop resistance against contemporary antimicrobial agents such as antibiotics. Infection with these pathogens can lead to prolonged hospital stays, higher medical costs and even increased patient mortality. Therefore, it is crucial to find new chemical compounds to help combat these pathogens. Plant-derived secondary metabolites characterized by easy availability and, in some cases, low toxicity and lack of harmful side effects may prove helpful in the fight against *Pseudomonas aeruginosa* and *Staphylococcus aureus*.

Objectives: This study aimed to investigate the bactericidal properties and toxicity of secondary metabolites found in selected plant species of *Helleborus* ssp.

Methods: Extracts were prepared from selected species of the *Helleborus* ssp. genus, i.e. *H. niger*, *H. viridis*, *H. purpurascens* using the following extraction methods: microwave-assisted water, methanol using ultrasonic waves, in tetrahydrofuran assisted desalting and using a Soxhlet apparatus. In addition, the extract with the highest activity was fractionated into columns. The antimicrobial potential of tested extracts was determined by Broth Microdilutions Method. The toxicity of antimicrobials was tested using the nematode *Caenorhabditis elegans* killing assay by determining its survival rate in the presence of analyzed concentrations of extracts

Conclusions: The performed experiments demonstrated the bactericidal effect of selected extracts. The most effective was the extract of *H. niger* in a Soxhlet apparatus using methanol as a solvent, which made it possible to obtain an extract having bactericidal activity against both pathogens tested. However, it was initially determined that each tested extract harmed nematode survival at lower concentrations than the minimum bactericidal concentration.

Keywords: *Helleborus*, Soxhlet apparatus, antimicrobial activity, nematodes

P-26: Antioxidant properties of *Polyscias filicifolia* (C. Moore ex E. Fourn.) L. H. Bailey (*Araliaceae*) callus extracts

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Polyscias filicifolia is native to South-Asia where is used in traditional medicine due to its adaptogenic, anti-inflammatory, antioxidant and antimicrobial activities. The phytochemical examinations of *P. filicifolia* shoots cultivated *in vitro* revealed the presence of oleane type saponins and phenolic compounds. Further the cytoprotective activity of its extracts was demonstrated. The aim of the study was to compare anti-oxidant potential of callus cultivated in presence of different growth regulators. The callus was initiated form leaves of *in vitro* growing plantlets and grow on solid MS medium supplemented with GA3 1 mg/L or GA3 3 mg/L and kinetin 1 mg/L under light/dark 12/12 h regime for 4 weeks. Next callus tissues were collected, lyophilized and extracted. Resulted extracts were investigated for their antioxidant properties including estimation of the content of total phenolics (TPC), total flavonoids (TFC) and the potential for scavenging of DPPH radical. Plant growth regulators had significant effect on TFC: in the culture variant supplemented together with GA3 and kinetin, TFC was over 2-fold higher than in variant containing only GA3, while TPC and DPPH radical scavenging activity were not affected.

Keywords: *P. filicifolia*, callus extracts, antioxidant properties

Culture variant	TPC [GA µg/g DW]	TFC [QE µg/g DW]	DPPH [%]
GA3 1 mg/L	231 ± 63,98	0,4 ± 0,19	86,8 ± 0,06
GA3 3 mg/L + kinetin 1 mg/L	233,5 ± 12,34	0,9 ± 0,58	87,4 ± 0,22

P-27: The nanofibrous mats for human cardiac cells culture

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Currently, numerous studies are conducted using nanofibers as a scaffold for culture cardiac cells however, there is still a lack of research evaluating the impact of physicochemical properties of polymer nanofibers on the functions of cardiac cells. This paper presents comparison and evaluation of the effect of two types of parallel arranged nanofibers made of polyurethane (PU) and poly(ε-caprolactone) (PCL) (Fig. 1A). Studies have been carried out to check how nanofibers affect human cardiomyocytes differentiated from induced pluripotent stem cells (hiPSC-CMs) and human cardiomyocytes (HCMs). High cell viability of cell cultures was noticed. Additionally, it was determined that hiPSC-CMs and HCMs showed a parallel arrangement and an elongated rod-like shape grown on nanofibers (Fig. 1B). The increased expression of Troponin T and α-actinin was noticed for the cells cultured on nanofibers mats (Fig. 1C). Based on the results, it can be concluded that human cardiac cells cultured on nanofibers leads to an increase in cell maturity in terms of their orientation, morphology and protein expression level compared with cells grown on polystyrene plates (PS).

Acknowledgments: *SONATA BIS 2019/34 / E / ST5 / 00381*

Keywords: tissue engineering, nanofibers, human cardiac cells, *in vitro* cardiac cell models

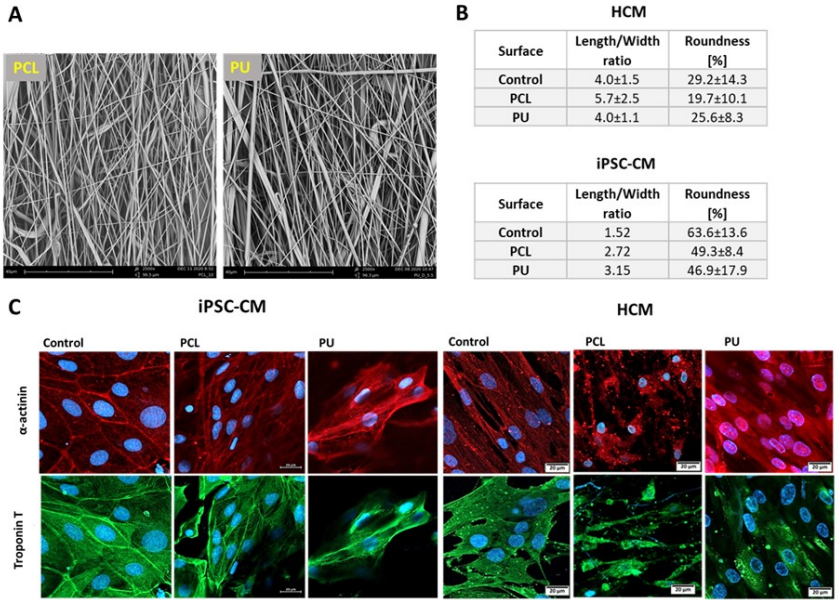

Fig. 1

P-28: Antarctic bacteria as a source of new compounds against anti-multidrug resistant microorganisms

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Antarctic bacteria can be a source of new biotechnologically important compounds. It was demonstrated that many pathogens are sensitive to some substances produced by psychrophiles.[1] *C. albicans* is a multi-drug resistant fungus that causes high mortality rate in immunocompromised and high-risk surgical patients.[2] The second group of microorganisms, which are considered as a threat in the area of human health comprises bacteria known as ESKAPE pathogens.[3] The purpose of this work was to check if selected strains of antarctic bacteria produce new antibiotics to which *C. albicans*, ESKAPE pathogens and other selected microorganisms are sensitive. Modified Cross-Streak Method was used to determine whether antarctic bacteria secrete compounds which inhibit growth of the tested microorganism. Twelve strains of pathogens were selected - one fungus (*C. albicans*) and eleven bacteria (*S. aureus*, *P. aeruginosa*, *K. pneumonia*, *S. typhimurium*, *E. coli*, *B. subtilis*). All tested strains of antarctic bacteria inhibited growth of *C. albicans* with varying strength. The appearance of other pathogens was not affected. Future research will focus on isolation, identification and separation of secreted by antarctic bacteria compounds. Such an approach may lead to the discovery of a new anti-*C. albicans* drug.

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Keywords: antibiotics, antarctic bacteria, ESKAPE, multi-drug resistant, *C. albicans*

P-29: Analysis of the effectiveness of anti-staphylococcal bacteriophage therapy in a human blood model

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The growing problem of antibiotic resistance of bacteria has become a global issue forcing us to look for an alternative therapeutic option to help treat infectious diseases effectively. In recent years, special interest was gained by bacteriophage therapy, the therapeutic use of phages, viruses that specifically infect bacteria in the treatment of multidrug-resistant bacteria. The immune system is an additional factor enhancing the effectiveness of the therapy allowing for the removal of harmful microbes. Unfortunately, knowledge concerning the immune system's response during bacteriophage therapy is limited. This study aimed to analyze the effectiveness of phage therapy and its impact on the phagocytic activity of neutrophils in a human blood model. The collected blood was prepared to obtain plasma and heat-inactivated plasma. The same amount of *S. aureus* and vB_SauM_A phage in different MOI was added to all samples. To determine the stability and therapeutic effectiveness of phages, the samples were incubated at 37℃. The phagocytic activity was analyzed according to the Wright method. In the next stage of research, *S. aureus* biofilm was used to determine the possibility of biofilm destruction by bacteriophages. After incubation, the models were stained with resazurin and crystal violet. The results showed normal infection and multiplication of phages in both types of plasma and no reduction in the number of active phages. In a biofilm model, the plasma and phages reduced biomass and viability, while thermal inactivation of plasma factors increased biomass. The treatment was highly effective in reducing *S. aureus* cells in a whole-blood model. The study suggests that bacteriophage therapy may be a future therapeutic alternative to antibiotics.

Keywords: Bacteriophage, Immunology

P-30: DNAJC30 gene variation in patients with Leber hereditary optic neuropathy

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Leber's hereditary optic neuropathy (LHON) is one of the first described mitochondrial diseases. It is manifested by bilateral, painless vision loss caused by degeneration of the perikaryons and axons of the retinal ganglion cells. Three main and several rare pathogenic variants in the mitochondrial genome (mtDNA) responsible for the occurrence of this disease have been detected. These variants are associated with dysfunction of respiratory chain complex I in the mitochondria. Although for many years mtDNA pathogenic variants were considered the only cause of the disease, they were not detected in many patients with clinically diagnosed LHON. Recently, dysfunction of the mitochondrial chaperone protein encoded by a nuclear DNAJC30 gene has been associated with the occurrence of the LHON phenotype. This chaperone is involved in the repair of subunits of the respiratory chain complex I. The aim of this study was to analyze the DNAJC30 gene coding sequence for pathogenic variants responsible for LHON as well as investigate the influence of the DNAJC30 gene variation and mtDNA variation on the appearance of the disease. The analysis was performed on DNA samples collected from three study groups: patients with an unexplained LHON genetic background, patients harboring a most common mitochondrial pathogenic variant m.11778G>A responsible for LHON and the asymptomatic carriers of that variant. Three potentially pathogenic variants were found in the coding sequence of the DNAJC30 gene, in the samples from the first study group. The most frequent was c.152A>G. This variant was found not only in all of the study groups, but also, although less often, in the control group. This finding supports the hypothesis of the occurrence of this variant as a consequence of the Eastern European founder effect, as proposed by Stenton *et al.* in 2021. The results from this study will have implications in terms of diagnosis and genetic counseling of LHON.

Keywords: *DNAJC30*, LHON, mtDNA, pathogenic variant

P-31: Evaluation of petroleum products biodegradation capabilities of Pseudomonas strains immobilized in alginate beads generated in microfluidic devices

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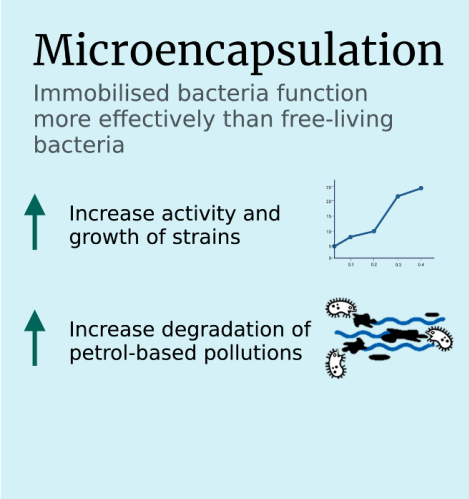
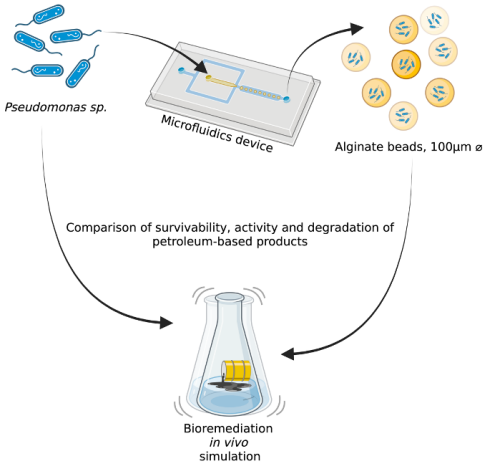
The aim of the study was to compare the hexadecane biodegradation capacity of different *Pseudomonas* strains. The comparison involved free-living and immobilized bacterial cells entrapped in alginate microcapsules produced using microfluidic methods. During the experiment, dehydrogenase activity and the numbers of microbes were measured weekly. At the end of the experiment, the hexadecane degradation rate was also analysed. The immobilized bacteria showed better growth, higher dehydrogenase activity and improved hexadecane degradation rate in comparison to the free-living bacteria. Taken together, these factors increased the survival rate of the tested strains and improved the degradation rate of contaminants. These results suggest that additional immobilization of microorganisms used in bioremediation technological processes can positively affect the adaptation phase, their growth rate and metabolic activity.

Keywords: alginate, biodegradation, bioremediation, hexadecane, immobilisation, microfluidic, microencapsulation, *Pseudomonas*, petroleum-based products





P-32: Effect of kappa opioid receptor activation on tumor cell proliferation in the mouse colon

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The involvement of kappa opioid receptors (KOP) in cancer development is not well understood. However, it is known that KOP receptors, which play a significant role in regulation of pain, anaesthesia, addiction and other physiological and pathological conditions, are essential for modulation of various cellular processes related to cancer. The objective of our study was to assess the impact of selective KOP receptor ligands on the colonic carcinogenesis. We carried out experiments on murine epithelial colorectal cancer cells (MC-38). MTT and LDH tests were used to assess how U50488H (a selective KOP agonist) and nor-Bni (a specific KOP antagonist) affect the cell viability and cytotoxicity. *In vivo*, U50488H was injected twice weekly at a dose of 1 mg/kg in the mouse model of colitis-associated colorectal cancer induced by azoxymethane and dextran sodium sulfate (AOM/DSS). We examined the mRNA expression of OPRK1 and PDYN in normal and colorectal cancerous tissue from mice. *In vitro*, U50488H reduced the cell viability after 24 hours, nor-Bni had no effect on the cell survival, but nor-Bni was able to offset the action of U50488H. *In vivo*, no discernible change in the colonic length between AOM/DSS and U50488H-treated group *in vivo*. Moreover, the colon was broader in U50488H as compared to AOM/DSS-treated group. When compared to the U50488H-treated group, the AOM/DSS-treated group displayed thicker submucosal and muscle layers as well as a rise in immune cell infiltration. In the distal colon the majority of the tumors were found in both groups, U50488H-treated group had more tumors than AOM/DSS-treated group. It was discovered that U50488H affected the progression of the colonic cancer.

Keywords: KOP, U50488H, nor-Bni, cancer, colon

P-33: Nanofibrous mats integrated with PC-PDMS-PC microsystem for hypoxia studies in cardiac cells

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Hypoxia plays a key role in cardiovascular pathologies and understanding its effects is essential to developing effective therapies. Our innovative PC-PDMS-PC microsystem offers a more efficient method for maintaining and studying hypoxia in cardiac cells. The microsystem enables simultaneous study of normoxia and hypoxia by regulating oxygen levels in cultured microchambers. One of the unique aspects of our system is the use of polyurethane (PU) nanofiber mats designed specifically for cardiac cell culture. These mats provide an ideal environment for long-term culture of rat cardiomyoblast cells (H9C2). Viability was assessed in normoxia for seven days, demonstrating the system's ability to maintain cultures for extended periods of time. To ensure accurate simulation of hypoxia and precise control of microsystem conditions, oxygen-sensitive SF-RPSu4 films were used. Oxygen concentrations in the hypoxia and normoxia zones were determined by adjusting the timing of nitrogen flow through the channels. After 60 minutes of nitrogen flow, oxygen concentrations reached 0% in the culture chambers. Importantly, exposure to hypoxia for five hours had a significant effect on H9C2 cells - ATP levels dropped by about 53%. In conclusion, the novel microsystem we developed with a nanofiber mat integrated with PC-PDMS-PC allows a comprehensive study of hypoxia and its effects on cells. This microsystem can achieve hypoxia within one hour and significantly affect ATP levels in H9C2 cells. As a result, this platform has the potential for future research on myocardial cell regeneration, providing an improved approach to studying hypoxia in cardiac cells using Lab-on-a-chip systems. By better understanding the effects of hypoxia on cardiac muscle cells, we can contribute to the development of more effective therapies and interventions for cardiovascular disease.

Keywords: hypoxia, microsystem, cardiac cells, nanofibrous mats

P-34: Synthesis, modification and application of magnetic nanoparticles in photothermal therapy

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Nowadays, as a result of the unique physical and chemical properties of magnetic nanoparticles, more scientists are getting interested in them. On this account, magnetic nanoparticles may be used in medicine, as a tool in diagnostics and treatments of cancer. The aim of conducted research was to apply magnetic core shell nanoparticles (Au@Fe2O3-PEG-OH and Au@Fe2O3-PEG-NH2) in anticancer therapy procedure. Magnetic nanoparticles can be applied in photothermal therapy (PTT) as photoactive agents. This kind of therapy use the ability of magnetic nanoparticles to convert the energy of electromagnetic radiation into thermal energy. According to that, as a result of irraditation of magnetic nanoparticles with near-infrared radiation, there is an increase in temperature, that leads to cell death. In the initial stage of research, after the synthesis of magnetic nanoparticles, their physical and chemical properties was checked (TEM, DLS, zeta potential, absorption spectrum). In the next stage, the energy conversion test was carried out. It confirmed that, the solutions of nanoparticles irradiated with electromagnetic radiation heated up to 76,3°C and 78,5°C, Au@Fe2O3-PEG-OH and Au@Fe2O3-PEG-NH2, respectively. Furthermore, the cytotoxicity of magnetic nanoparticles was analyzed. All research was conducted on four human cell lines (cancer and normal cell lines). For non-toxic concentrations of nanoparticles solutions, the PTT procedure was performed.

Keywords: magnetic nanoparticles, photothermal therapy, anticancer therapy

P-35: MLK4 mutations in metastasis

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Mixed-Lineage Kinase 4 (MLK4) is frequently overexpressed in several types of cancer. Previous studies have suggested that MLK4 could regulate its downstream targets, such as ERK, JNK, p38 and NF-κB signaling pathways. Moreover, MLK4 has been found as a novel, frequently mutated gene and a driver of metastases. However, further research is needed to fully understand the functional consequences of MLK4 mutations and their effects on downstream signaling pathways. In this study, we aim to uncover the MLK4 interactome and changes in the MLK4 binding partners caused by somatic mutations, thereby providing a comprehensive view of the possible involvement of MLK4 in the mechanism of metastases. We first used the site-directed mutagenesis to generate mutations in the HA-tagged MLK4 vector and then transiently transfected MLK4 vectors into HEK293T cells. Our results showed that three mutations in the kinase domain significantly decreased the phosphorylation of ERK and JNK. In contrast, the level of phosphorylation of NF-κB was higher in mutants than in MLK4 WT, providing the regulation of the NF-κB pathway by MLK4. Subsequently, we observed the decreased level of Snail, a mesenchymal marker and stable level of E-cadherin, an epithelial marker, in all studied mutations in the HCC1806 CRISPR-MLK4 knock-out breast cancer cell line. Next, we used coimmunoprecipitation (co-IP)-mass spectrometry to assess the MLK4 interactome and the effects of specific mutations on the changes of those interactions in HCC1806 cancer cells. In our preliminary results, we identified the binding partners of MLK4 related to cell cycle, metabolism, scaffold, chaperone, heat shock, nucleotide-binding protein. In future research, we will continue to investigate the MLK4-interactome and the changes of these interactions caused by MLK4 mutants, therefore comprehensively characterizing the molecular details and functional significance of MLK4 mutations and their interactions in metastasis.

Keywords: MLK4, mutation, interaction, metastasis

P-36: Application of modern biotechnological, phytochemical and molecular techniques to obtain secondary metabolites from *in vitro* cultures of *Rindera graeca* (A. DC.) Boiss. & Heldr. (*Boraginaceae*) transgenic roots

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In vitro cultures of plant cells, tissues and organs constitute a part of biotechnology, which importance grows in production of natural compounds for use in medicine, cosmetics and food industry. Plant biotechnology methods allow to cultivate only organs or tissues directly related to biosynthesis of these metabolites. From an array of methods used in plant biotechnology, genetically and biochemically stable transgenic roots, characterized by a high proliferative potential are increasingly used in bioactive compound production. *Rindera graeca* is an endemic plant growing on rocky slopes of Greek mountains. Chemical composition of *R. graeca* is not yet fully known, however it has been recently reported that transgenic roots of this species can be a source of newly discovered naphthoquinone – rinderol, a compound characterized by a high pro-apoptotic potential towards tumorous cells. Further these roots produce significant amounts of phenolic acids with dominating lithospermic acid B, these compounds play an important role in the prevention of civilization diseases. The goal of the studies was to select hairy root lines characterized by the highest biosynthetic potential of secondary metabolites from naphthoquinone group and phenolic compounds with anticancer, antimicrobial and anti-inflammatory properties. Fourteen cell lines of transformed *R. graeca* roots grew in 250 mL flasks containing 50 mL of DCR medium for 4 weeks. Next, roots were collected, freeze-dried and extracted with n-hexane. Dry extracts were analyzed using HPLC-DAD method. Analysis showed that most of the transgenic root lines produced naphthoquinone dyes, mainly rinderol. The highest concentrations of rinderol were found in R4 (900.58 µg/g DW) and R2 (116.01 µg/g DW) lines while the highest concentration of rinderol in the medium were found in the cultures of R3 (30.09 mg/L) and R7 (30.73 mg/L) cell lines.

Keywords: *In vitro* culture, *R. graeca*, riderol, hairy roots, trangenic roots

P-37: Effect of locomotor exercise on the density of Vesicular Glutamate Transporter 2 distributed in the ventral horn of the lumbar spinal cord in rats with spinal cord injury

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VGluT2 is a transporter that takes up glutamate into synaptic vesicles in neurons and serves as a marker of glutamatergic input in the spinal network, including spinal interneurons and some descending inputs (Broadhead *et al.* Sci Rep., 2020). Complete spinal cord transection (SCT) leads to the disruption of these inputs, resulting in downregulation of VGluT2 transcripts (Ziemlińska *et al.*, PLOS One, 2014). This study aimed to investigate whether SCT in the low thoracic Th13 segment results in a decrease in VGluT2 protein distribution in the ventral horn gray matter, medial part (Rexed laminae VII-VIII, rich in interneurons) and lateral part (lamina IX, rich in motoneurons) and whether locomotor exercise may be effective in increasing VGluT2 levels. Experiments were conducted on 3 groups of adult rats: intact (C, n=6), spinal (Sp, n=8) and spinal subjected to treadmill locomotor training, supplemented with tail stimulation (SpLoc, n=8) performed in 7 x 3 min sessions, for 5 weeks. Evaluation of the hindlimb stepping was assessed by two independent researchers based on videos taken weekly. Six weeks after SCT we observed over 4-fold increase of the number of step-like movements in SpLoc compared to Sp rats. The training significantly improved the quality of locomotion, manifested by higher number of plantar foot placements and step alternations. Immunohistochemical analysis of VGluT2 optical density was performed on lumbar L5 sections. Light microscopy images were collected and subjected to the analysis using the ImagePro Plus 7.0 software. Surprisingly, after SCT, despite the changes in VGluT2 mRNA level, we observed no changes in VGluT2 protein signal in the medial or lateral part of gray matter. Locomotor training did not change VGluT2 protein level either. Our results suggest that observed functional changes are not related to VGluT2 transporter abundance in the L5 lumbar network.

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Keywords: spinal cord, VGluT2, motoneurons, injury

P-38: Targeting PD-1/PD-L1 immune checkpoint, different profiles of druggability between human and mouse PD-L1

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Moving into pre-clinical *in vivo* evaluation of anticancer properties of PD-1/PD-L1 inhibitors requires the creation of an immunologically appropriate model. When choosing the appropriate *in vivo* model for testing immune checkpoint inhibitors, two aspects have to be considered: (1) murine or full reconstitution of the human immune system; (2) the differences within the PD-L1 sequence and tertiary structure between hPD-L1 mPD-L1 that may cause different druggability profiles of the proteins. So far, in the research on newly developed potential inhibitors both syngeneic mouse models as well as the models based on immunocompetent knock-in-mice with fully human or mouse/human hybrids of immune checkpoints were introduced 4. In recent years immunodeficient mouse with the recapitulated human immune system and the engraftment of human cancer cell lines or PDXs has been announced as a versatile model for preclinical immunotherapeutic testing. As the main limitations of these models are costs and time consumption, there is a need to provide a reliable platform for rapid screening of PD-L1 inhibitors' ability to *in vitro* interference with both hPD-1/hPD-L1 and hPD-1/mPD-L1 immune checkpoint. Thus, the goal of the research was to develop a new cell-based assay for mechanistic verification of the inhibitors' ability to reactivation of immune response as the effect of blockade of hPD-1/mPD-L1 immune checkpoint and its use for the immunostimulatory characterization of both newly developed PD-L1 inhibitors and mAbs used in clinical therapies, for which no data has been presented so far. What is more, the obtained results have been enriched with the interdisciplinary studies using NMR spectroscopy, HTRF assay and crystallography.

Keywords: cancer, immunotherapy, immune response, immune checkpoint, PD-1/PD-L1 interaction

P-39: Characterization and potential applications of *Yersinia enterocolitica* phages in minced meat

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Yersinia enterocolitica is a foodborne pathogen that is commonly found in raw meats. It is a significant cause of gastroenteritis, especially in children. Antibiotic resistance in *Y. enterocolitica* strains is a growing concern, highlighting the need for alternative control methods such as bacteriophages – viruses that infect bacteria. In this work, we characterized *Y. enterocolitica* phages and investigated their potential application in minced meat. Three phages: YEP1, YEP2 and YEP3 were isolated from sewage samples collected from a pig farm. The phages were propagated using *Y. enterocolitica* as the host bacteria showing high specificity for this species, with no activity against other tested bacteria. Bacteriophages were also characterized by whole genome analysis. We conducted challenge tests using artificially contaminated meat samples to evaluate the application of *Y. enterocolitica* phages in minced meat. The samples were inoculated with *Y. enterocolitica* and treated with the phages at different concentrations. The results showed that YEP1 and YEP2 reduced the bacterial load by approx. 85% units, while YEP3 reduced it by 50%. The phages were effective even at low concentrations, with a minimum effective concentration of 10^3 PFU/g of meat. In conclusion, this work highlights the potential of *Y. enterocolitica* phages as an effective control method for *Y. enterocolitica* in minced meat. The phages showed high specificity and were effective even at low concentrations. Further research is needed to optimize the application of the phages in meat products and to evaluate their safety and efficacy under different storage and processing conditions. Nonetheless, our findings offer promising insights for the development of phage-based strategies to improve food safety and reduce the risk of *Y. enterocolitica* infections in humans.

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Keywords: *Yersinia enterocolitica*, bacteriophages, food safety, phage product

P-40: Study of the effect of selected saponins on the transport of cytostatic drugs into cells

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Saponins are a group of glycosides of natural origin. They show the ability to reduce surface tension in aqueous solutions. Due to their foaming properties, they have found application in the cosmetic industry. Saponins, thanks to their ability to penetrate the lipid layer of the cell membrane, can alter its permeability. The purpose of this study was to determine the effect of the presence of saponins on the efficiency of the introduction of therapeutic substances into cells using the electroporation method. Electroporation (EP) is a process during which temporary pores are formed in the cell membrane under the influence of an electric field of appropriate voltage. These pores provide a pathway for the penetration of substances into the cells. In the first stage of the study, the cytotoxicity of four selected saponins (escin, digitonin, QBS and glycyrrhetinic acid) was evaluated using the MTT assay. The study was conducted on four cell lines: skin (cancerous A375 and normal HaCaT) and lung (cancerous A549 and normal MRC-5). For each saponin, 2 non-toxic concentrations were selected and used in subsequent stages of the study to evaluate the efficiency of electroporation. The efficiency of saponin-assisted electroporation was determined by evaluating the penetration of propidium iodide (PI) into the cells. This dye has no ability to penetrate into living cells whose membranes are intact. During electroporation, when pores are created in the membrane, the dye enters the cells. Thus, the use of PI during conducted electroporation allows us to evaluate the effectiveness of this method. Based on the results obtained, it was found that the presence of saponins increases the efficiency of electroporation and this effect is different for the cell lines used in the study. It was observed that the efficiency of PI penetration into the cells of the A375 line is 9 times higher for electroporated samples than when the samples were only incubated with the escin solution.

Keywords: saponins, electroporation, transport of cytostatic

P-41: Comparison of antimicrobial properties of selected dairy cow udder care preparations based on propolis extracts

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Mastitis is a common condition that occurs in dairy cows. It is caused by environmental and infectious pathogens. Udder inflammation causes many economic and breeding losses as well as reduces technological value of milk. Nowadays, because of increasing antibiotic resistance of pathogens strains, researchers are looking for new alternative methods to prevent mastitis with compositions enriched with natural ingredients such as propolis. The aim of the study was to compare the antimicrobial properties of different preparation from 5 fabricants. For the experiment milk from cows which was diagnosed with mastitis was used. The SCC parameter was over 400 thousand/ml. To multiply a strains of pathogens, culture was performed on specialized selective media such as Edwards Lab Agar with the addition of bovine blood, Mannitol Salt Lab Agar, Chromogenic Candida Lab Agar (Biomaxima, Lublin, Poland) and Chromogenic Uri-Color Lab Agar and Selective Medium for the Isolation of Prototheca according to methodology Pore *et al.* 1973. The Incubation lasted 24 h in 37 °C. The cultured microorganisms (gram negative bacteria, gram positive bacteria, algae and fungi), *Prototheca* spp. were suspended in 0.9% NaCl solution. and the suspension was pipetted into a 96-well plate. In each plate 50μl of solution and 50μl of 5 commercially available preparations were places and a control sample was included. The plate was then incubated for 24h at 37 °C. After 24h, 10μl of XTT reagent was added to the wells and incubation was continued for another 24h. After time, the absorbance was read spectrophotometrically and the statistical analyses were performed. Comparing the 5 preparations and analyzing their composition, it can be concluded that the most valuable biocidal effect is shown by product No. 5. Composition, form and also density has an impact for the antimicrobial properties. The pathogen that proved most resistant to the biocidal effect was *S. aureus*. The most sensitive was *S. uberis*.

Keywords: antimicrobial, mastitis, pathogens, proplis extracts, cow

P-42: The optimization of the immobilization process of bacteriophage T4 on a carrier based on bacterial cellulose

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Bacteriophages are viruses that replicate within bacteria and are used to search for new methods of combating pathogenic bacteria. Currently, these specific viruses are considered as an alternative to antibiotics. Bacteriophage T4, which infects *Escherichia coli*, was used for research. On LB agar plates with the addition of the host bacteria (*Escherichia coli*), bacteriophage T4 was added, which had previously been incubated in bacterial cellulose for 24 hours. The effect of the size and method of extraction of bacteriophage in bacterial cellulose was examined. All factors tested affected immobilization efficiency. The best results were obtained for bacterial cellulose before drying, by precipitating bacteriophages from it in a NaCl solution. In the next stage, it is planned to investigate whether the structure of bacteriophage affects immobilization efficiency in bacterial cellulose.

Keywords: microbiology, bacteriophage, bacterial cellulose

P-43: Survival Analysis of *Tenebrio Molitor* L. Beetles Injected with Neuropeptides under Thermal Stress Conditions

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Insects make up the largest group of animals and can thrive in various environments and temperatures, owing to their exceptional ability to endure a wide range of environmental stressors, such as extreme temperatures. It has been observed that insects' exposure to cold stress affects various physiological processes. The neuroendocrine system, along with its associated mediators, such as neuropeptides and biogenic amines, is known to regulate insects' physiological and behavioral processes and thus, could potentially influence their thermal tolerance. To investigate this *Tenebrio molitor* beetles that were 30 days old were exposed to one hour of heat stress at 40°C and cold stress at –5°C. The objective of this investigation was to determine whether the injection of neuropeptides could influence the survival of insects over a 21-day observation period. In the study, four neuropeptides were employed: tachykinin-related peptides (TRPs), which are substances that are related to the overall response of the insect organism to stressors; short neuropeptide F (sNPF), which is responsible for feeding; allatostatin type C (PISCF), which is a metabolically active peptide; and proctolin, a potent myostimulator. These neuropeptides were administered following exposure to both heat and cold stress.

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Keywords: cold stress, insects, neurohormones, neuropeptides, survival rate