



SYMBIOZA

INTERNATIONAL BIOTECHNOLOGY
SYMPOSIUM

The 13th Prof. Krzysztof W. Szewczyk
International Biotechnology Symposium

SYMBIOZA

BOOK of ABSTRACTS

8–10 May 2026, Warsaw

The 13th Krzysztof W. Szewczyk
International Biotechnology Symposium "Symbioza"
Book of Abstracts
8–10 May 2026, Warsaw.
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Warsaw, 8 May 2026.

Dear Participants,

it is an honour and a true joy to welcome you to the 13th International Biotechnology Symposium ‘Symbioza’. With each passing year, this event becomes more than just a conference. This thirteenth edition, perhaps the luckiest one yet, stands as a testament to the dedication, passion and hard work of many remarkable and talented individuals who have contributed their time and energy to bring this initiative to life. Behind this conference are countless hours of work, ideas, challenges, and shared moments that brought us here. I am deeply proud of every single person who contributed to making this event a reality.

What began over a decade ago as an initiative of students from three Warsaw universities, has grown into a dynamic and thriving community. Today, the Warsaw Society of Biotechnology ‘Symbioza’ brings together not only students of Warsaw, but also members from scientific institutions across all Poland. This growth reflects the strength of our shared division: to connect people, ideas and opportunities within the field of biotechnology. It is not only something to celebrate, but also a responsibility we carry with great care. To continue creating a space that is open, inclusive, and meaningful.

‘Symbioza’ has always been more than just a conference. It is a space where you can share your research, sometimes for the very first time, and be heard. A space where you can listen to inspiring lectures from internationally recognized experts and realize that science is not distant, but within reach. A space where you can meet people working in academia, industry, and beyond, and begin to understand the many paths that lie ahead of you. But above all, it is a space for people. A space to meet those who think like you, who question like you, who are driven by the same curiosity and ambition. A space where conversations over coffee can turn into collaborations, friendships, or even future careers. A space where you can exchange ideas, challenge perspectives, and grow, not only as scientists, but as individuals.

As organisers, we have always believed in building this space primarily for students, by students. Every edition is created with the intention of responding to a constantly evolving scientific world, while staying true to our core mission: to connect, to support, and to inspire. I encourage you to make the most of this occasion: ask questions, share your ideas and take inspiration from one another.

Thank you for being part of this journey and welcome to the 13th International Biotechnology Symposium ‘Symbioza’!

On behalf of the organising committee



— **Maria Kalenik**,

President of the Warsaw Society of Biotechnology ‘Symbioza’

Honorary Patron of the Symposium

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 Interfaculty 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.

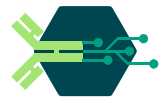
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Knowledge partner



Sponsors

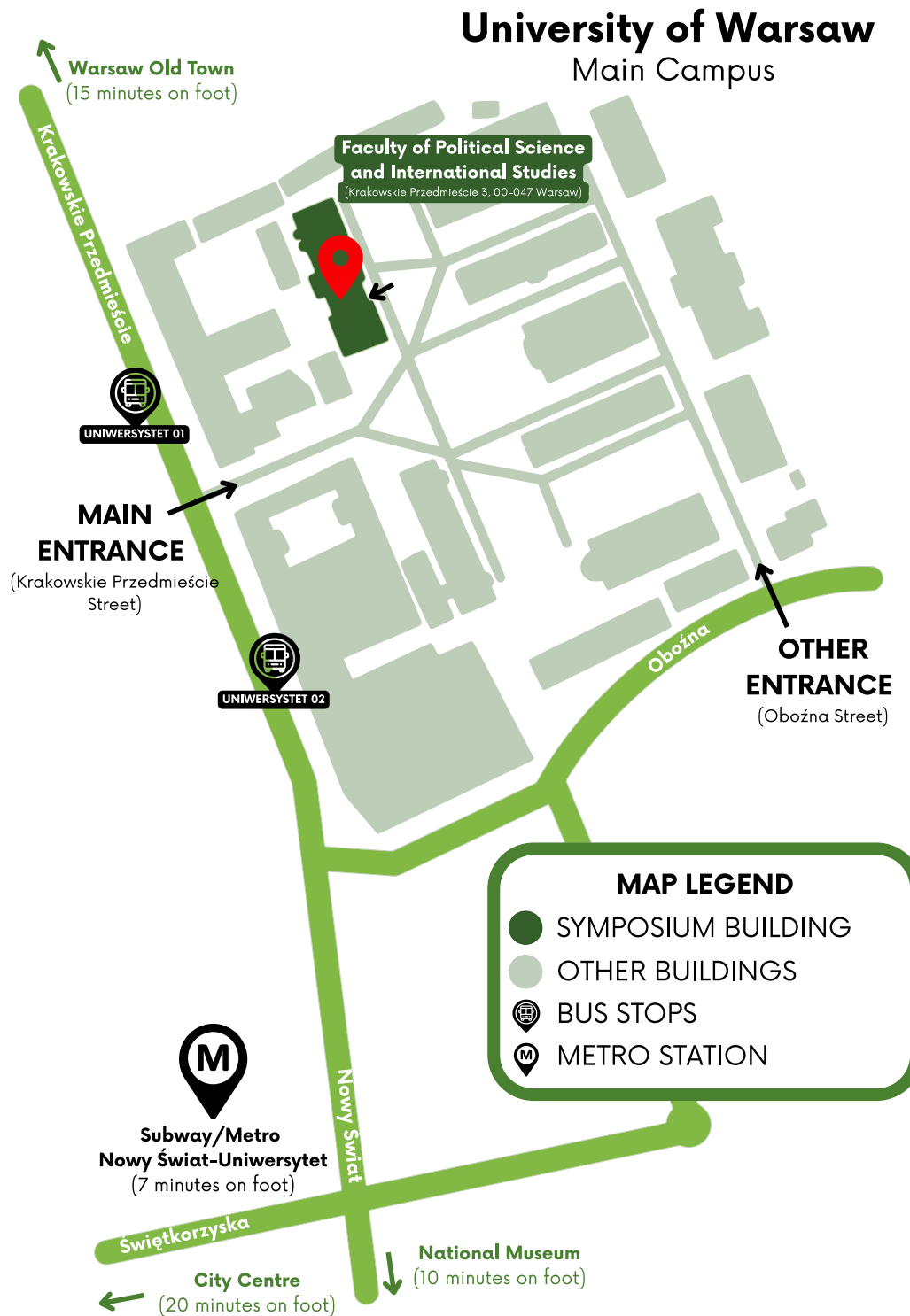


Media patronage



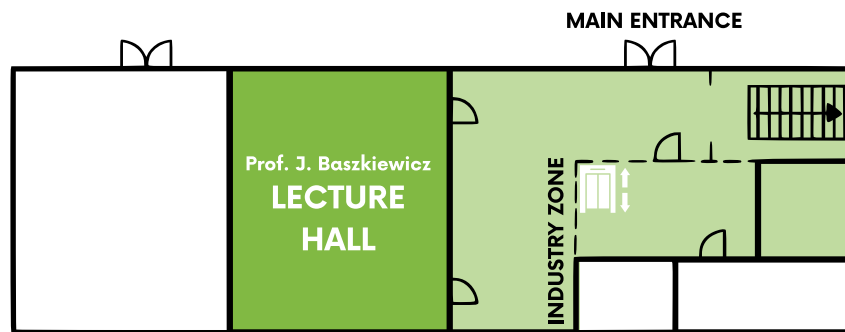
Conference Venue

Campus Map



Conference Building

The Faculty of Political Science and International Studies (WNPiSM), University of Warsaw, is located on the university’s Main Campus. Although political science is a relatively modern academic discipline, its roots at the university date back to 1917, and the Faculty has been housed in its current building since 1976. Today, it serves as a leading research hub comprising 16 departments. Research areas encompass political science, international relations, internal security, social policy, and political philosophy. Additionally, WNPiSM hosts numerous prominent academic initiatives and research units, including the Shevah Weiss Center for Israel Research and the Jewish Diaspora, the Center for Research on Social Security, the Tadeusz Mazowiecki Chair, and the Human Trafficking Research Center.



Floor 0



Floor +3

- LECTURE, WORKSHOPS, AND SESSIONS ROOMS
- CONFERENCE SPACE
- ELEVATORS
- RESTROOMS

Organiser



SYMBIOZA WARSAW SOCIETY OF BIOTECHNOLOGY

Warsaw Society of Biotechnology “Symbioza” (WSB “Symbioza”) was founded in 2013 through the collaborative efforts of students from the University of Warsaw, Warsaw University of Life Sciences, and Warsaw University of Technology. Its mission is to cultivate a platform for the exchange of knowledge and experiences among biotechnology students and researchers. At the heart of WSB “Symbioza”, there is our flagship event: the Internatitonal (earlier: Intercollegiate) Biotechnology Symposium “Symbioza” (IBS “Symbioza”), which you are currently attending. Held annually from 2012, the Symposium serves as a platform for international students and PhD candidates to present their research findings and engage with peers and experts in the field. Recognized for its excellence, it was honoured as the “Conference of the Year 2019” in the prestigious StRuNa Competition.

Beyond academic conferences, WSB “Symbioza” hosts initiatives for biotechnology enthusiasts such as *Symbioza Umysłów (Symbiosis of Minds)* or *OAK Attractive Conventicles Camps*. *Symbiosis of Minds* is directed to polish high school students interested in biotechnology. *Symbiosis of Minds* mainly focuses on demonstrating that science is a unity, with all its fields interconnecting. In the current global scientific research, increasing emphasis is placed on projects that bridge different, often seemingly distant disciplines.

During *OAKs*, 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.

With its diverse range of activities and commitment to promoting biotechnology awareness and education, WSB “Symbioza” continues to inspire and empower the next generation of life sciences leaders, fostering a global network of collaboration and innovation.

Organising Committee

Committee Leaders

Aleksandra Sijka
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Magdalena Karasek
Maja Sadowska

Maria Krasuska
Zuzanna Opalska
Zuzanna Sulej

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Zofia Izdebska — Vice-President, Head of External Communication
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Iga Jancewicz, PhD
4Cell Therapies

Tomasz Kamiński, PhD
University of Warsaw

Robert Stryński, PhD
University of Warmia and Mazury, Olsztyn

Co-organisers

The Symposium has been organised by the members of the Warsaw Society of Biotechnology “Symbioza” annually since 2013. Acknowledging its own roots, every year the Society invites life sciences-oriented student organisations of the host city to participate in shaping the event. The 2026 supporting organisers are:

Biotechnology Student Association “Herbion” — 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 — 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’ Society for Microbiology — a student organization at the University of Warsaw affiliated with the Department of Medical Microbiology (formerly the Department of Applied Microbiology at the University of Warsaw) since its inception. The Society was established in 2003, and its founder and first president was Tomasz Jagielski (currently the academic supervisor of the Society). The main goal of the Society is to promote knowledge in the field of microbiology and related disciplines. This goal is pursued through the organization of seminars, discussion panels, and scientific sessions. Meetings are conducted by its members or invited guests—specialists, researchers, and practitioners. In addition, members of the Society participate in independent experimental projects, organize scientific workshops for primary and secondary school students, and take part in study visits to various research centres both in Poland and abroad.



Program overview

DAY 1 | FRIDAY, MAY 8th

13:00-15:00	Registration	17:20-17:40	Coffee break
15:00-15:15	Opening Ceremony	17:40-18:30	Plenary lecture <i>João Marques Garcia</i>
15:15-16:05	Plenary lecture <i>Tomasz Kościółek</i>	18:30-18:40	Coffee break
16:05-17:20	Discussion panel	18:40-20:40	Workshops

DAY 2 | SATURDAY, MAY 9th

09:20-10:20	Oral sessions #1-2	13:00-13:50	Plenary lecture <i>Anna Schulten</i>
10:20-10:30	Coffee break	13:50-15:40	Lunch
10:30-11:20	Plenary lecture <i>Carlos Perea Resa</i>	15:40-16:40	Oral sessions #5-6
11:20-11:40	Coffee break	16:40-18:10	Poster session
11:40-12:40	Oral sessions #3-4	18:10-19:00	Plenary lecture <i>Emilio Mármol Sánchez</i>
12:40-13:00	Coffee break	19:00-21:00	Social event

DAY 3 | SUNDAY, MAY 10th

10:00-11:00	Oral sessions #7-8	13:20-14:00	Brunch
11:00-11:50	Plenary lecture <i>Zuzanna Krysiak</i>	14:00-15:15	Discussion panel
11:50-13:20	Poster session	15:15-15:45	Closing Ceremony

Agenda

Friday, 8 May 2026

13:00 – 15:00 **Registration**

15:00 – 15:15 **Opening Ceremony** (**Baszkiewicz Hall**)

15:15 – 16:05 **Plenary Lecture** (**Baszkiewicz Hall**)

PL-1 *Is the gut microbiome the future of precision medicine?*

TOMASZ KOŚCIÓŁEK, *Sano Centre for Computational Medicine, Krakow (PL)*

16:05 – 17:20 **Discussion Panel** (**Baszkiewicz Hall**)

D-1 *At the Crossroads of Science: Interdisciplinarity in Biotechnology*

PANELISTS: *Lucja Kowalewska, Jan Guzowski, Lukasz Andrzejewski*

17:20 – 17:40 **Coffee Break**

17:40 – 18:30 **Plenary lecture** (**Baszkiewicz Hall**)

PL-2 *Tackling Cultivated Meat Challenges at WUR through Research and Education*

JOÃO MARQUES GARCIA, *Wageningen University and Research Center (NL)*

18:30 – 18:40 **Coffee Break**

18:40 – 20:40 **Workshops** (**Rooms 303 & 317**)

W-1 *Microbiology in Practice*

ALEKSANDRA WICHROWSKA, *Bacteromic (PL)*

W-2 *Science commercialisation – how to turn scientific ideas into real-world solutions*

MICHAŁ JESKA, *Medical Innovation Institute (PL)*

Saturday, 9 May 2026

9:20 – 10:20 **Parallel oral session: “Catch Me If You Can”** (Baszkiewicz Hall)

O-1 Hijacking Host Immunity: How *Fasciola hepatica* Challenges Vaccine Development

ALICJA LASKOWSKA, *University of Warsaw (PL)*

O-2 Parasite-derived extracellular vesicles regulate NLRP3 inflammasome signalling in an *in vitro* model of intestinal inflammation

MAGDALENA STAWICKA, *University of Warmia and Mazury, Olsztyn (PL)*

O-3 Cross-species differences in dsRNA detection and translation shutdown among influenza virus hosts

KAROLINA PIANKA, *University of Warsaw (PL)*

O-4 Proxypylline derivative versus classical calcineurin inhibitors: a novel approach for controlling cytokine storms in SARS-CoV-2 infection

KSENIYA GIENKA, *Warsaw University of Technology (PL)*

9:20 – 10:20 **Parallel oral session: “Split Happens”** (Room 303)

O-5 Toward a Genetically Modifiable Neutrophil Model: Functional Assessment of CEBP α -Induced Differentiation in K562 Cells

SALEM HAMDAN, *Medical University of Warsaw (PL)*

O-6 Human Induced Pluripotent Stem Cell-Derived Cardiomyocytes and Cardiac Progenitor Cells for Cell-Based Therapy in Murine Model of Myocardial Infarction

KAROLINA ŁASZCZ, *Jagiellonian University, Kraków (PL)*

O-7 Bone marrow endothelial cells as phagocytes: uptake of stressed erythrocytes and microparticles

MATEUSZ SAR, *Jagiellonian University, Kraków (PL)*

O-8 Sfl is translationally controlled through Igf2bp2 to regulate hematopoiesis

DANIEL GRYGOROWICZ, *International Institute of Molecular Mechanisms and Machines, Polish Academy of Sciences, Warsaw & University of Warsaw (PL)*

10:30 - 11:20 **Plenary lecture** (Baszkiewicz Hall)

PL-3 Preservation and recycling of protein complexes across mitosis: molecular memory

CARLOS PEREA RESA, *Centro de Biología Molecular Severo Ochoa, Madrid (ES)*

11:20 – 11:40 **Coffee Break**

11:40 – 12:40 **Parallel oral session: “Survival of the Smallest”** (Baszkiewicz Hall)

- O-9** Evolutionary pressure observed *in vitro* – a case of *Pseudomonas aeruginosa* bacteriophages developing lytic host adaptation
ALEKSANDRA ZALEWSKA, *University of Gdańsk (PL)*
- O-10** Microfluidic system for long-term imaging of optically thin bacterial biofilms to correlate phenotypic heterogeneity with local microenvironment
KLAUDIA STAŚKIEWICZ, *Institute of Physical Chemistry PAS, Warsaw (PL)*
- O-11** The structural and protective role of myxoxanthophyll in stabilizing the photosynthetic apparatus of cyanobacteria
KAMIL DRZYMAŁA, *Jagiellonian University, Kraków (PL)*
- O-12** Susceptibility of environmental *Pseudomonas* isolates to P2D1 tailocins reveals potential impacts on microbial populations beyond Soft Rot *Pectobacteriaceae*
KACPER TOMASIK, *University of Gdańsk (PL)*

11:40 – 12:40 **Parallel oral session: “Fantastic Nanos and Where to Find Them”** (Room 303)

- O-13** Synthesis of gold nanoparticles modified with a dinucleotide cap analog as potential biomedical carrier
ALEKSANDRA KOPER, *University of Warsaw (PL)*
- O-14** Adapting an IMC foundation model for prediction of clinical features
KACPER PIETRZYK, *University of Warsaw (PL)*
- O-15** Fe₃O₄@AuNPs@Au@Pt nanozymes as promising multifunctional agents for biomedical diagnostic and anticancer applications
MONIKA ŚMIGIELSKA, *Warsaw University of Technology (PL)*
- O-16** Reductase-mimicking nanozymes for novel signal generation strategy in paper-based immunoassays
PAWEŁ STAŃCZAK, *Warsaw University of Technology (PL)*

12:40 – 13:00 **Coffee Break**

13:00 – 13:50 **Plenary lecture** (Baszkiewicz Hall)

- PL-4** How do plants remember winter? Insights from a cold-induced epigenetic switch
ANNA SCHULTEN, *Department of Cell and Developmental Biology, John Innes Centre, Norwich (UK)*

13:50 – 15:40 **Lunch** (3rd Floor)

15:40 – 16:40 **Parallel oral session: “Fast and folded”** (Baszkiewicz Hall)

O-17 Purification of human poli(A) polymerase using ÄKTA liquid chromatography system
ZOFIA PIETRZAK, Adam Mickiewicz University, Poznań (PL)

O-18 Identification of new components of the lipoprotein secretion system in *Porphyromonas gingivalis*
WERONIKA PIETRAS, Jagiellonian University, Kraków (PL)

O-19 “The LES(s) I know, the better” – characterisation of lipoprotein export signal in the unique secretion system of *Porphyromonas gingivalis*
ALEKSANDRA CZERKOWICZ, Jagiellonian University, Kraków (PL)

O-20 Unlocking non-canonical rescue of the ubiquitin-proteasome system under functional compromise
KAROLINA MILCZ, International Institute of Molecular and Cell Biology in Warsaw (PL)

15:40 – 16:40 **Parallel oral session: “The Devil Wears Antigens”** (Room 303)

O-21 Deep Learning in tumor immune microenvironment analysis from multiplex imaging
JAKUB WINIARSKI, University of Warsaw (PL)

O-22 High-Sensitivity Sequential cfDNA Testing of MDM2 and HSP90AB1 Copy Levels for Progression Monitoring in Soft Tissue Sarcoma Patients
WIKTORIA TRYNKOS, Warsaw University of Technology (PL)

O-23 *In vitro* studies on the anticancer properties of graphene oxide aerosol, using pancreatic ductal adenocarcinoma cells as a biological model
ALEKSANDRA CIECHOŃSKA, Warsaw University of Life Sciences (PL)

O-24 Microfluidic-Assisted Precise Bioprinting of Hydrogel Micro-Bioreactors
LEON JURKIEWICZ, Institute of Physical Chemistry Polish Academy of Sciences, Warsaw (PL)

16:40 – 18:10 **Poster session 1: P-1–P-49** (3rd Floor)

18:10 – 19:00 **Plenary lecture** (Baszkiewicz Hall)

PL-5 From ancient DNA to ancient RNA and beyond: Toward a multi-omics future in paleogenetics

EMILIO MÁRMOL SÁNCHEZ, Center for Evolutionary Hologenomics, The Globe Institute, University of Copenhagen, Copenhagen (DK)

19:00 – 21.00 **Social Event** (Conference Building)

21:00 – **Conference Party** (Bolek Pub & Restaurant)

Sunday, 10 May 2026

10:00 – 11:00 **Parallel oral session: “Catabolism, Anabolism & Other Bad Life Decisions”** (Baszkiewicz Hall)

O-25 Enzymes of microsomal fractions of *Arabidopsis* leaves, involved in the synthesis of esters of free fatty acids with primary alcohols – biochemical characterisation and substrates specificity

ALICJA CZYŻ, *University of Gdańsk & Medical University of Gdańsk (PL)*

O-26 Fabrication of Laser-Induced Graphene Electrodes for Lactate Biosensing

IPEK SARIER, *Warsaw University of Technology (PL)*

O-27 Influence of Medium pH on *In vitro* Growth, Vegetative Propagation and Phytochemical Properties of *Drosera capensis* L.

KONRAD ADAM MICHALIK, *Medical University of Warsaw (PL)*

O-28 Cytotoxic activity of geranyl resveratrol derivatives in cutaneous squamous cell carcinoma models

NADIA FATYGA, *Ludwik Rydygier Collegium Medicum, Bydgoszcz & Nicolaus Copernicus University, Toruń (PL)*

10:00 – 11:00 **Parallel oral session: “H₂O: Just a Drop is Enough”** (Room 303)

O-29 Impact of silver nanoparticles and ions on development and maturation of guppy (*Poecilia reticulata*)

JAN PRUSZYŃSKI, *Warsaw University of Life Sciences (PL)*

O-30 The presence of linezolid-resistant *E. faecium* and *E. faecalis* strains in wastewater treatment plant environments

JULIA CZUBA, *Ludwik Rydygier Collegium Medicum, Bydgoszcz & Nicolaus Copernicus University, Toruń (PL)*

O-31 Is the Cure for Neuroblastoma Hidden in the Baltic Sea?

MONIKA ZIELENKIEWICZ, *University of Gdańsk (PL)*

O-32 HydroBioCell – development of an innovative, biodegradable, and antimicrobial water filter based on bacterial cellulose

ALINA ZWOLIŃSKA, *West Pomeranian University of Technology, Szczecin (PL)*

11:00 – 11:50 **Plenary lecture** (Baszkiewicz Hall)

PL-6 Advanced polymer nanomaterials for regenerative medicine and light-activated therapy

ZUZANNA KRYSIAK, *Department of Biosystems and Soft Matter, Institute of Fundamental Technological Research, Polish Academy of Sciences, Warsaw (PL)*

11:50 – 13:20 **Poster session 2 (P-50–P-95) (3rd Floor)**

13:20 – 14:00 **Brunch (Ground Floor)**

14:00 – 15:15 **Discussion panel (Baszkiewicz Hall)**

D-2 Science in Action: From Education to Media

PANELISTS: *Katarzyna Siuzdak, Katarzyna Koziak, Marta Mielcarek*

15:15 – 15:45 **Closing Ceremony & Prizes (Baszkiewicz Hall)**

Poster Session 1

Saturday, 9 May, 16:40 – 18:10

- P-1** Identification of Novel Natural Compound Derivatives Modulating Large-Conductance Calcium-Activated Potassium (BKCa) Channels
ANGELIKA SKÓRA, *University of Warsaw (PL)*
- P-2** Modulation of the TRPA1 channel affects functional properties of human pancreatic stellate cells in a phenotype-dependent manner
JAKUB WOJTAS, *Jagiellonian University, Kraków (PL)*
- P-3** Antifungal activity of a gemini surfactant against *Aspergillus flavus* isolated from feed
KINGA KOSZELA, *Lodz University of Technology (PL)*
- P-4** Assessment of biofilm formation by *Escherichia coli* 8917 on common 3D-printing polymers: applications in biosensor and bioreactor design
OLIWIA JANISZEWSKA, *University of Gdańsk (PL)*
- P-5** Bridging the Gap: Optimizing Direct Reprogramming of Human Fibroblasts into Neurons for Morphological Analysis
JULIA ŁUKASZEWICZ, *University of Warsaw & International Institute of Molecular and Cell Biology, Warsaw (PL)*
- P-6** Establishing a FingR-based system for imaging PSD-95 in NT2-derived neurons
ANNA KUSIAK, *Jagiellonian University, Kraków (PL)*
- P-7** Evaluation of Cellular Stress-Related Proteins in the Cerebral Cortex of Trap1-Mutant Male Mice: A Novel Model of ASD
ZUZANNA SALWOCKA, *University of Warsaw (PL)*
- P-8** Development and validation of a bioinformatic pipeline for Ion Torrent sequencing data in forensic SNP analysis
KATARZYNA URBANELIS, *Jagiellonian University, Kraków (PL)*
- P-9** Intracellular Complement Components Regulate Metabolism and Stress Response in Bone Marrow Stromal and Hematopoietic Cells
DAMIAN WOJDALSKI, *Medical University of Warsaw & Warsaw University of Technology (PL)*
- P-10** The effect of leaf extracts from *Pistacia vera* and *Rhus glabra* in combination with ionizing radiation on normal and cancerous breast cells
SZYMON SZYMCZYK, *University of Łódź (PL)*
- P-11** The TRPA1 channel differentially regulates calcium signalling in phenotypically distinct human pancreatic stellate cells
AGNIESZKA RYŁKO, *Jagiellonian University, Kraków (PL)*

- P-12** Uncovering Selective Anticancer Potential of Cyanobacterial Metabolites from *Nostoc edaphicum* CCNP1411 Against Neuroblastoma
WIKTORIA JELENIEWSKA, *University of Gdańsk (PL)*
- P-13** Enhancing the nutritional profile of pasta dough through microencapsulated polyphenols
WERONIKA BIŃKOWSKA, *Warsaw University of Life Sciences (PL)*
- P-14** TheraChip – development of a simulation-supported microfluidic lab-on-a-chip for 3D spheroid culture for personalized cancer photodynamic therapy
LENA PIOTROWSKA, *Warsaw University of Technology (PL)*
- P-15** Computational design of Dengue virus NS2B/NS3 protease inhibitors
ADAM BORYS, *University of Gdansk (PL)*
- P-16** Illuminating parasite immunology: Successful bacterial expression of a fluorescent *Toxocara canis* cystatin
MATEUSZ STELMASIAK, *Warsaw University of Life Sciences (PL)*
- P-17** Hemp meal as an alternative to fish meal in European perch (*Perca fluviatilis*) aquaculture
WIKTORIA CIEŚLA, *Warsaw University of Life Sciences (PL)*
- P-18** Sensitivity of *Pseudomonas donghuensis* P482 to sodium nitroprusside- and S-nitrosoglutathione-induced nitrosative stress is influenced by the growth medium
JAN JEZIEWSKI, *University of Gdańsk & Medical University of Gdańsk (PL)*
- P-19** Methicillin Resistance and SCCmec Diversity in *Staphylococcus pseudintermedius* Isolated from Dogs
RAFAŁ NEJFELD, *Warsaw University of Life Sciences (PL)*
- P-20** Accelerated prototyping of recombinant proteins using a transient expression system in *Nicotiana tabacum* BY-2 cell packs
JAKUB KORNIATOWSKI, *Medical University of Warsaw (PL)*
- P-21** How to Deliver a Peptide Across the Skin? A Case Study of BPC-157
JULITA KOSTKA, *Lodz University of Technology (PL)*
- P-22** Sensor Arrays for Rapid Genetic Point-of-Care Diagnostics: Optimization of DNA Probes for Bacterial Identification
BIBIANA MICHALSKA, *Warsaw University of Technology (PL)*
- P-23** Cellular response to DNA damage induced by 405 nm laser irradiation
ALEKSANDRA RZECZYC, *Jagiellonian University, Kraków (PL)*
- P-24** Electrochemical tongue – a sensor array for the detection of date-rape drugs (DRDs) and other pharmaceuticals
JOANNA KOPROWSKA, *Institute of Physical Chemistry, Polish Academy of Sciences, Warsaw (PL)*

- P-25** When intracellular calcium is not enough: extracellular influx drives T Cell activation
KATARZYNA GONERA, *University of Gdańsk (PL)*
- P-26** Alginate Hydrogel-Based Delivery of Antimicrobial Peptides: Controlled Release and Preserved Antibacterial Activity
MICHAŁ IDZIAKOWSKI, *University of Warsaw (PL)*
- P-27** RyhB1 and RyhB2 sRNAs shape Iron-Dependent Regulatory Networks in *Yersinia enterocolitica*
PAULINA LIPSKA, *University of Warsaw (PL)*
- P-28** Effects of use of Foliar Silica on growth of *Vitis vinifera* L. Grapevine
MARGARET HOPE NYAMBURA, *AGH University, Kraków (PL)*
- P-29** Characterization of a Newly Isolated Bacteriophage 4.1 Targeting *Salmonella enterica*
KINGA MALINOWSKA, *University of Gdańsk (PL)*
- P-30** Integrated mRNA and miRNA profiling of the hermaphroditic Hgy3 cucumber line
ERYK WENERSKI, *Warsaw University of Life Sciences (PL)*
- P-31** Verification of the effectiveness of magnetic nanoparticle-mediated photothermal therapy in a lab-on-a-chip system
DARIA MAJEWSKA, *Warsaw University of Technology (PL)*
- P-32** Redefining the biosynthetic potential of *Aralia spinosa* L. – establishment and phytochemical profiling of hairy root cultures
MACIEJ OBREŃBSKI, *Medical University of Warsaw (PL)*
- P-33** Screening of the *Pseudomonas donghuensis* P482 miniTn5 mutant library for its ability to form biofilms on abiotic surfaces and plant tissues
EWA ŚPIEWAK, *University of Gdansk & Medical University of Gdansk*
- P-34** The role of interleukin 6 in the replication kinetics of selected poxviruses in mouse dendritic cells *in vitro*
ZUZANNA MACHERZYŃSKA, *Warsaw University of Life Sciences (PL)*
- P-35** Biochar Functionalized with Rejet Water-Derived Humic Substances as a Sustainable Biostimulant and Protective Agent for Rapeseed against *Botrytis cinerea*
BENIAMIN BEDNARZ, *University of Silesia, Katowice (PL)*
- P-36** Integrated approach to biotechnological regeneration of Na-P1 zeolitic sorbent contaminated with diesel oil
WERONIKA PILECKA, *University of Warsaw (PL)*
- P-37** Polyphenol-coated hybrid magnetic nanozymes for combined photothermal and chemodynamic anticancer therapy
JAN GÓRNIASZEK, *Warsaw University of Technology (PL)*

- P-38** Dissecting Hair Follicle Stem Cells Niche Dynamics at Single-Cell Resolution through BMP signaling Modulation
AJAY JAKHAR, *University of Warsaw (PL)*
- P-39** Optimization of expression and purification of recombinant West Nile Virus NS2B-NS3 protease
KARINA FRANCHUK, *University of Gdansk & Medical University of Gdansk*
- P-40** Data-driven calibration of collective event detection in spatio-temporal cell signaling
ADELA ŻELACHOWSKA, *University of Warsaw (PL)*
- P-41** Effects of *Bacillus*-Derived Osmoprotectants on Enzymatic Activity and Microbial Communities Composition in Saline Soils
ANNA BERNATOWICZ, *University of Warsaw (PL)*
- P-42** Biotransformation of *Hypericum perforatum* compounds by selected probiotic products
GABRIELA ZEGAN, *Medical University of Warsaw (PL)*
- P-43** Overcoming Microbiological Instability: Preservation Techniques for Spent Coffee Grounds as a Sustainable Substrate for Biotechnological Applications
ANNA MALICKA, *Warsaw University of Life Sciences (PL)*
- P-44** Mechanism of action of new 4,5,6,7-tetrabromo-1H-benzimidazole derivatives in breast cancer
ANIELA KUBIAK, *Warsaw University of Technology (PL)*
- P-45** Cell line-dependent microRNA expression in canine osteosarcoma *in vitro* models
JAKUB PODOBA, *Wroclaw University of Environmental and Life Sciences (PL)*
- P-46** Polysaccharide nanocarriers functionalized with targeting and chelating ligands for theranostics
MARTYNA CYGAN, *Lodz University of Technology (PL)*
- P-47** Effects of light intensity on the growth of selected duckweed species (*Lemna minor*, *Landoltia punctata*, and *Spirodela polyrhiza*)
PIOTR CICHY, *Silesian University of Technology, Gliwice (PL)*
- P-48** The effect of silver nanoparticles on *Toxoplasma gondii* tachyzoites
IGA MARINKOVIĆ, *University of Warsaw (PL)*
- P-49** Structural and sequence determinants of efficient nonconventional splicing in *Euglena longa*: a comparison of constitutive and alternative introns
OLIWIA ZALEWSKA, *University of Warsaw (PL)*

Poster Session 2

Sunday, 10 May, 11:50 – 13:20

- P-50** Small heroes, great power: bacterial metabolites as sustainable alternatives for the farms of the future
JUSTYNA WRONKA, *University of Warsaw (PL)*
- P-51** Differentiation-Dependent Effects of ATP and Serotonin on Neuronal Cell Survival
WIKTORIA PIÓRKOWSKA, *Ludwik Rydygier Collegium Medicum Nicolaus Copernicus University, Bydgoszcz & Nicolaus Copernicus University, Toruń (PL)*
- P-52** Lineage Tracing Reveals the Contribution of MCAM-Positive Plastic Tumor Cells to Breast Cancer Progression
MEGHA GAUTAM, *University of Warsaw (PL)*
- P-53** Optimising mass of biochars from diverse feedstocks for humic substance recovery from two types of reject water
JUSTYNA MICHALSKA, *Silesian University of Technology, Gliwice (PL)*
- P-54** Design and Expression of Recombinant Mitochondrial Proteins in FreeStyle™ 293-F Cells
JUSTYNA JAZOWSKA, *University of Warsaw (PL)*
- P-55** AgNPs from ascorbic acid-rich plant waste: sonochemical synthesis and antimicrobial activity
ANNA NALEPA, *Cracow University of Technology (PL)*
- P-56** Biotechnological Potential of Fabaceae *In Vitro* Cultures for Phenolic Compound Production with Cosmetic Applications
OLEKSANDRA LABAN, *University of Szczecin (PL)*
- P-57** Targeting replication stress: MK-8776 and disulfiram trigger apoptosis in ovarian cancer with cell line-specific kinetics
WIKTORIA BĘBENEK, *University of Lodz (PL)*
- P-58** *In silico* analysis of transcription factor binding sites of plastoglobule-related genes in response to abiotic stress in the *Arabidopsis thaliana* model
TOMASZ BACHLEDA-GRÓBARZ, *University of Warsaw (PL)*
- P-59** Combined Surface-Enhanced Raman Spectroscopy And Chemometric Analysis Of Blood Plasma And Bone Marrow For Rapid Detection And Monitoring Of Pediatric Acute Lymphoblastic Leukemia
PATRYCJA WIEWIÓROWSKA, *Institute of Physical Chemistry, Polish Academy of Sciences (PL)*

- P-60** Green synthesis of gold nanoparticles using milk thistle seed extract and their cytotoxicity toward skin cells
ZOFIA NOWAK, *Warsaw University of Life Sciences (PL)*
- P-61** Affinity of plant Rab proteins for REP protein
WIKTORIA WINDAK, *Warsaw University of Life Sciences (PL)*
- P-62** Selection and characterization of transgenic *Arabidopsis thaliana* plants with knocked out gene encoding the mitoribosomal protein Rps23
ALEKSANDRA WACHOWSKA, *University of Wroclaw (PL)*
- P-63** Event-Based Laser Speckle Imaging for Real-Time Assessment of Microbial Activity Across Varying Inoculum Densities
EDUARDS TEODORS MINCIS, *University of Latvia (LV)*
- P-64** *Aralia spinosa* L. hairy roots as a biotechnological platform for araloside A production: optimization of *in vitro* culture conditions, growth kinetics, and biosynthetic efficiency assessment
RAFAŁ KIELKIEWICZ, *Medical University of Warsaw (PL)*
- P-65** Plant responses to persistent pollutants: bioaccumulation and stress effects of perfluoroalkyl substances in *Solanum dulcamara in vitro*
NATALIA TOŁOCZKO, *University of Gdansk (PL)*
- P-66** Enhancing the Specificity of DNA Methylation Monitoring through Effective Removal of Non-Specifically Adsorbed HRP: A Comparative Approach
SANU K ANAND, *Warsaw University of Technology (PL)*
- P-67** Comparative Genomic Analysis and Experimental Validation of Genetic Variants in Five Cucumber (*Cucumis sativus* L.) Lines
JAN SZTENKE, *Warsaw University of Life Sciences (PL)*
- P-68** Plant-based expression systems: A novel platform for producing recombinant therapeutic proteins
ANTONINA KAWKA, *Medical University of Warsaw (PL)*
- P-69** IFIT1 oligomerization kinetic study
JOANNA GRZYMKOWSKA, *Warsaw University of Technology (PL)*
- P-70** Comparison of the effect of fetal bovine serum (FBS) and bovine calf serum (BCS) on the development of ferroptosis resistance in the HT-1080 fibrosarcoma cell line
DOMINIKA BIAŁEK, *Warsaw University of Life Sciences (PL)*
- P-71** C-methylated polytrimethylenimines: synthesis, antimicrobial properties, and the role of molecular mass
ALEKSANDRA KOWALSKA, *Warsaw University of Technology (PL)*
- P-72** The impact of JAO2 gene mutation on secondary growth and xylem formation in the *Arabidopsis thaliana* stem: anatomical and gene expression analyses
JULIA STĘPNIAK, *Adam Mickiewicz University, Poznań (PL)*

- P-73** Regulating MMP-9-Dependent Neuronal Plasticity In Situ
JULIA ZAREMBA, *University of Warsaw (PL)*
- P-74** Selenium biofortification of Chinese cabbage using a native plant growth-promoting bacterial consortium
TATSIANA CHERVANETS, *University of Warsaw (PL)*
- P-75** Long-read metabarcoding and single-cell genomics: uncovering protist diversity in Masurian peatlands
KAROLINA KRUPA, *University of Warsaw (PL)*
- P-76** Anti-leukemic action of new protein kinase CK2 inhibitors
WIKTORIA KĘSIK, *Warsaw University of Technology (PL)*
- P-77** Identification of *Ochrobactrum anthropi* ATCC49188 genes essential for the colonization of biotic and abiotic surfaces
ZIEMOWIT JUSZCZUK, *University of Gdansk (PL)*
- P-78** *Aegeritella superficialis* symbiosis – antagonistic or mutualistic? New light on the ant-infecting fungus 50 years after species description
ZOFIA BULANDA, *University of Warsaw (PL)*
- P-79** *In vitro* model of early symptoms of diabetic nephropathy: hyperglycemia induced rapid changes to mitochondrial respiration and biogenesis in renal tubular cells
MARTYNA OSTROWSKA, *Mossakowski Medical Research Institute, PAS (PL)*
- P-80** N-Methylated Insulin-Derived Peptides as Inhibitors of β -Amyloid Aggregation: A Potential Strategy for Targeting Neurodegenerative Diseases
PATRYCJA GRABARCZYK, *Lodz University of Technology (PL)*
- P-81** Transcriptome-wide mapping of m6A sites in MDA-MB-231 cells
SZYMON ŁUKASZEWICZ, *Adam Mickiewicz University, Poznań (PL)*
- P-82** Phytohormones as a Novel Therapeutic Concept in the Treatment of Triple Negative Breast Cancer (TNBC)
IRENA WADAS, *Warsaw University of Life Sciences (PL)*
- P-83** Potential of Environmental Yeast Strains in Winemaking
MARIUSZ KRYSIAK, *Warsaw University of Life Sciences (PL)*
- P-84** Label-Free Electrochemical Monitoring of Microcystin-LR Release from *Microcystis aeruginosa* Using an Electrified Liquid-Liquid Interface
MUTHAIAH ANNALAKSHMI, *University of Łódź (PL)*
- P-85** Impact of Purification Method on the Anti-Inflammatory Activity and Green Chemistry of Aspirin Synthesis
GABRIELLA BIENIEK, *Coventry University Wrocław (PL)*

- P-86** Enhancing Serological Immunoassay Performance via Nanozyme-Functionalized and Microfluidic Systems
KAROLINA PORZYCKA, *Warsaw University of Technology (PL)*
- P-87** Photomodulated effect of graphene oxide and chlorophyll-based substrates on the morphological and functional status of U87 glioma cells *in vitro*
IVAN VYSOTSKYI, *Warsaw University of Life Sciences (PL)*
- P-88** From Surface to Sensor: Optimizing Screen-Printed Electrodes for Biosensing Applications
ALEKSANDRA SKIBA, *Warsaw University of Technology (PL)*
- P-89** Tracking the drop: Investigating erythroid ACKR1 in the Duffy-null neutrophil mystery
NIKOLA SOBCEK, *Ludwig-Maximilians-Universität, Munich (DE)*
- P-90** Graphene Quantum Dots Modulate Photosynthetic Pigments Without Affecting Growth in *Lepidium sativum*
ECE NUR ERDIM, *Ordu University (TR)*
- P-91** *Pontechium maculatum* (L.) Böhle&Hilger as a model for *in vitro* culture establishment and conservation of a steppe relict species
MARIA ROSIAK, *Wroclaw University of Environmental and Life Sciences (PL)*
- P-92** Bioinformatic analysis and optimization of recombinant HuFABP7 protein production in *Pichia pastoris*
MAGDALENA KARASEK, *University of Warsaw (PL)*
- P-93** The effect of CHK1 kinase inhibitor and aldehyde dehydrogenase inhibitor on OVCAR8 ovarian cancer cells
MAGDA GRANOSIK, *University of Łódź (PL)*
- P-94** Structural and functional characterization of pco and sil copper resistance regions in *Cronobacter sakazakii*
ALEKSANDRA CAĐDEREK, *University of Warsaw (PL)*
- P-95** Impact of NETs on mitochondrial function in ECTV-infected cells – preliminary study
POLA PRUCHNIAK, *Warsaw University of Life Sciences (PL)*

PL-1: Is the gut microbiome the future of precision medicine?

Tomasz Kościółek^{*,1}

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The human gut microbiome plays a central role in host physiology, contributing to digestion, metabolism, and immune regulation. It is increasingly recognized as a promising therapeutic target, with interventions ranging from dietary modulation and probiotics to fecal microbiota transplantation and live biotherapeutics.

Despite substantial progress, the traditional ecological framework used to associate microbiome composition with host phenotypes is reaching its limits. While marker gene (16S rRNA) sequencing has long served as a cost-effective and informative standard, the field is undergoing a transition toward shotgun metagenomics, which provides comprehensive access to the microbial gene repertoire along with functional insights and strain-level resolution.

In this work, we leverage advances in shotgun metagenomics together with modern machine learning to develop predictive and interpretable models of the gut microbiome. We explore function prediction using deepFRI, highlighting the advantages of homology-independent approaches that incorporate structural information and enable broad coverage of microbial genes. Building on this, we introduce GUT-FORMer, a foundation model framework that integrates taxonomic and functional signals into a unified representation of the microbiome.

Together, these approaches demonstrate the potential of combining large-scale metagenomic data with representation learning to improve microbiome-based inference. I will present recent results illustrating how such models can support robust diagnostic strategies and provide new insights into host-microbiome interactions, with implications for personalized interventions.



Dr. Tomasz Kościółek is a head of the Structural and Functional Genomics research team at the Sano Centre for Computational Medicine in Kraków, Assistant Professor at the Faculty of Computer Science of the AGH University of Kraków, co-founder and Chief Scientific Officer of onebiome Sp. z o.o., member of the Polish Academy of Sciences' Young Academy (term 2024–29), and Board Member of the Polish Bioinformatics Society for the 2026–28 term. He obtained his Ph.D. in Biological Sciences from University College London (UK), and completed a postdoctoral fellowship in Rob Knight's group at the University of California San Diego (USA). From 2019 to 2023, he led a bioinformatics group at the Małopolska Centre of Biotechnology, Jagiellonian University in Kraków, and from 2023 to 2024 he was a university professor in the Department of Data Engineering and Exploratory Data Analysis at the Silesian University of Technology in Gliwice. He has received grants from NAWA, NCN, FNP and NCBR. He is the author of over 30 peer-reviewed scientific publications, including in *Nature*, *Nature Biotechnology*, and *Nature Communications*, and is a co-inventor on three patent applications. He works on developing and applying computational methods to better understand the function and dynamics of the human gut microbiome. He contributed to the development of widely used microbiome analysis tools, QIIME 2 and Qiita, and also works on state-of-the-art machine learning and statistical methods for protein function prediction (deepFRI) and for modeling dysbiosis and microbiome dynamics. The goal of his group is to build a multi-level understanding of the microbiome - from genes, through structures, to functions and therapies.

PL-2: Tackling Cultivated Meat Challenges at WUR through Research and Education

*João Marques Garcia**¹

¹ Wageningen University and Research Center (NL)

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The transition toward a more sustainable and ethical food system demands innovative approaches to protein production. Cultivated meat and seafood have emerged as a compelling solution, yet significant scientific and technological barriers remain between laboratory-scale cell culture and commercially viable manufacturing. The Cellular Agriculture team within the Bioprocess Engineering group at Wageningen University & Research (WUR) is addressing these challenges through an integrated research program spanning the full production pipeline, from cell line development and bioprocess scale-up to final product formulation. Active projects include the scale-up of cultivated fish fat processes, the development of octopus muscle cell lines as a novel model organism, and the engineering of sustainable, animal-free culture media using microalgae hydrolysates as amino acid sources. The team is also advancing plant-based microcarrier technology to support anchorage-dependent cell growth at scale. Underpinning all experimental work is an integrated bioprocess design framework that combines cell and bioreactor modelling with techno-economic and environmental assessments, enabling systematic evaluation of process innovations before costly scale-up. Alongside research, WUR is pioneering cultivated meat education in Europe, offering the first dedicated master-level course on cultivated meat and seafood production.



Dr. João Marques Garcia is a biomedical engineer and holds a PhD on tissue engineering and regenerative medicine. Between 2022 and 2024, Joao joined the Directorate of Human and Robotic Exploration at the European Space Agency as a research fellow. Here, he explored the feasibility of using cultivated meat to feed astronauts in long-term space missions. As of March 2024, Joao was appointed Assistant Professor at the Wageningen University, where he is involved in research on cellular agriculture, and cultivated meat and seafood. In particular, Joao is interested in the development of robust cell lines, optimization of culture media formulations, and scaffold development. In addition, Joao is the coordinator of the master course on Cultivated Meat and Seafood, the first of its kind in Europe.

PL-3: Preservation and Recycling of protein complexes across mitosis: molecular memory

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Mitosis is not only essential for maintaining cell numbers in multicellular organisms but also provides a privileged window for resetting gene expression programs and generating cell diversity during development. Our laboratory investigates how gene expression is regulated through mitosis, focusing on the preservation and recycling of transcriptional machinery and RNA molecules during this transition. We have uncovered that the cohesin complex, traditionally known for its role in chromosome segregation, is also maintained through mitosis and plays a crucial role in ensuring accurate transcriptional reactivation in daughter cells. This function of cohesin appears essential for establishing tissue-specific gene expression profiles and reveals new molecular insights into the etiology of Cornelia de Lange syndrome. Building on this, we are exploring how the core transcriptional apparatus, particularly RNA polymerase II and its associated factors, is recycled after mitosis. Our studies aim to uncover how this process supports asymmetric stem cell division and how its gradual loss contributes to age-related transcriptional decline. Finally, our most recent line of research investigates the stability and transmission of RNA molecules across mitosis, examining how these preserved transcripts may contribute to gene expression memory and the acquisition of aberrant traits during cell transformation in cancer. Together, these projects shed light on the continuity of gene expression control through cell division and its broader implications for development, aging, and disease.



Dr. Carlos Perea Resa is a Junior Principal Investigator at CBMSO-CSIC, funded by the Ramón y Cajal program. His research focuses on transcription regulation, RNA biology, and cell cycle control, with the aim of understanding how gene expression is reprogrammed during development and in response to stress, particularly through mitosis and its impact on cell fate decisions. He currently leads a research line aimed at deciphering the molecular mechanisms governing the transmission of transcriptional machinery through mitosis and its role in cell differentiation. His long-term goal is to define fundamental principles of gene regulation during development and contribute to the understanding and treatment of developmental disorders. Previously, he investigated the role of cohesin in transcriptional repression during mitosis, providing new insights into the molecular basis of Cornelia de Lange Syndrome. At Massachusetts General Hospital-Harvard Medical School, he demonstrated that cohesin contributes to the global shutdown of transcription during mitosis and may participate in transcriptional reactivation after cell division. Across his career, he has developed broad expertise in gene expression regulation using diverse model systems, including mammalian cells, *Xenopus*, and *Arabidopsis*. He has built a strong foundation in molecular and cellular biology, particularly in transcription, RNA biology, and cell cycle regulation, and has established an international research network. He is now focused on consolidating his independent research program at CBM and contributing to the Spanish research system.

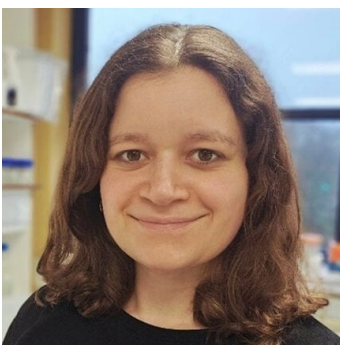
PL-4: How do plants remember winter? Insights from a cold-induced epigenetic switch

Anna Schulten^{*,1}

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Plants are masters at integrating environmental cues to regulate their growth and development. But how are such environmental signals translated into changes in gene expression? We use the floral repressor FLOWERING LOCUS C (FLC) in *Arabidopsis thaliana* to investigate cold-induced epigenetic gene silencing during vernalization, a process in which prolonged winter cold promotes flowering. This silencing is mediated by Polycomb-dependent repressive histone modifications at FLC, which require VEL proteins that assemble into multimeric complexes through dynamic head-to-tail polymerization. We dissect the role of VEL polymerization domains in vivo, revealing their combinatorial roles in maintaining the chromatin association of Polycomb proteins to facilitate the switch to epigenetic silencing. Our findings shed light on the mechanisms of environment-responsive gene silencing and highlight the broader relevance of higher-order protein assemblies in eukaryotic gene regulation.



Dr. Anna Schulten is a senior postdoc in the lab of Prof. Dame Caroline Dean at the John Innes Centre in Norwich (United Kingdom), where she studies the mechanisms underlying epigenetic silencing in the model plant *Arabidopsis thaliana*. The floral repressor gene FLC serves as a key example, undergoing epigenetic silencing in response to winter cold to ensure that flowering aligns with spring. Anna obtained her PhD from the Ruhr-University Bochum in Germany, where she investigated the function of a transcription factor involved in plant copper homeostasis. In September 2026, she will start her own research group as a junior group leader at CEITEC MU in Brno (Czech Republic), investigating the mechanisms underlying epigenetic memory in plants.

PL-5: From ancient DNA to ancient RNA and beyond: Toward a multi-omics future in Paleogenetics

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Ancient DNA has revolutionized our understanding of extinct and extant organisms, enabling the reconstruction of lost ecosystems and their long-gone inhabitants. However, our knowledge of ancient biology has been largely constrained to information derived from short DNA fragments or low-resolution protein sequences. While these have yielded remarkable insights, DNA primarily captures the static genetic code, and proteins merely provide a partial view of the coding fraction of the genome.

Trapped in between lies RNA, a molecular intermediate long dismissed as too transient and unstable to survive far beyond cell death. Recent breakthroughs in isolating, sequencing, and characterizing historical and ancient transcriptomes have challenged this assumption, revealing that RNA preservation may be more common than previously recognized.

This keynote will trace the troubled lifetime of ancient RNA as a research field, present recent advances enabling transcriptome recovery from historical and archaeological specimens of iconic extinct species such as the Tasmanian tiger and the woolly mammoth, and describe how ancient RNA is transforming our understanding of gene regulation, developmental biology, and infectious disease through time. Looking forward, I will discuss ongoing projects, future directions, and the unrealized potential of RNA and other biomolecules toward a multi-omics framework aimed at decoding the functional and structural biology of the past.



Dr. Emilio Mármol Sánchez is a research fellow in the Center for Evolutionary Hologenomics at the University of Copenhagen. Trained as a veterinarian and bioinformatician, he began his research career in Barcelona by studying gene regulatory networks and RNA biology in domestic animals. After completing his PhD in 2020, he transitioned into the emerging field of ancient transcriptomics, where during his postdoctoral work in Stockholm he developed novel laboratory and computational approaches for the isolation, sequencing, and computational analysis of ancient and historical RNA from extinct species. Now based in Copenhagen, his research is focused on a multi-omics approach to develop novel methods applied to historical and ancient animal specimens, spanning short- and long-read DNA sequencing, RNA profiling, chromatin reconstruction, epigenetic inference, and spatial deconvolution. Through such integrative framework, his work aims to reconstruct the functional biology of the past using molecular technologies of the present.

PL-6: Advanced polymer nanomaterials for regenerative medicine and light-activated therapy

*Zuzanna Krysiak**¹

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Advanced polymer nanomaterials are playing an increasingly important role in modern regenerative medicine, drug delivery, and light-activated therapies. These materials combine biodegradable polymers, nanofibers, hydrogels, and functional nanoparticles to create smart biomedical systems capable of interacting with biological tissues at the cellular level. Polymeric nanostructures can be engineered to deliver drugs in a controlled manner, support tissue regeneration, and respond to external stimuli such as light or temperature. Electrospun polymer nanofibers are particularly important because they mimic the structure of the natural extracellular matrix, making them suitable for tissue engineering and regenerative medicine applications. Hydrogels and crosslinked polymer networks can provide a moist, biocompatible environment for cell growth and drug delivery. The incorporation of nanoparticles, especially metallic nanoparticles such as gold nanostructures, enables photothermal effects, allowing light-activated therapy and controlled drug release. Light-responsive nanomaterials are a promising approach for minimally invasive therapies, enabling on-demand drug release via near-infrared light. Such systems may improve treatment efficiency while reducing side effects. Overall, advanced polymer nanomaterials represent a rapidly developing field that combines materials science, nanotechnology, and medicine, offering new possibilities for regenerative therapies, targeted drug delivery, and smart biomedical devices.



Dr. Zuzanna Krysiak obtained her PhD in 2022 from AGH University of Science and Technology in Kraków, Poland, where she investigated electrospun fiber-based skin patches for atopic skin. She is an Assistant Professor in the Division of Functional Polymer Nanomaterials, led by Filippo Pierini, at the Institute of Fundamental Technological Research, Polish Academy of Sciences. She has several years of experience in the design and manufacture of biomaterials for atopic skin treatment, wound healing and scaffolds, gained in domestic and international research centres. Experience in research projects based on electrospun fibers, hydrogels, and intelligent structures combining these two types of materials, confirmed by numerous publications and two patents. Knowledge of techniques for isolating human and animal cells, as well as culturing primary cell lines. Currently, she is focused on a light-responsive platform for miRNA delivery for wound healing.

D-1: At the Crossroads of Science: Interdisciplinarity in Biotechnology

The biggest innovations in biotechnology happen at the intersection of different scientific fields - and thus, need cooperation of scientists from diverse backgrounds, as well as an inspiring leader who can push collaboration in the right direction and blend multiple perspectives into one, cohesive goal.

During this discussion panel, we will debate various aspects of this topic, as well as the challenges it presents, amongst them successfully transforming innovative ideas into actual projects and applying an interdisciplinary approach in everyday operations. As group leaders, our panelists tackle these issues - and much more - on a daily basis, working in the sectors of fundamental and applied research, as well as in business development. The audience will have the unique opportunity to get inspiration and insight from different perspectives and contribute by asking questions during the Q&A session.

The discussion will be moderated by **Kacper Koźluk**, who will create the environment for open and helpful exchange of experience, asking the important question and steering the conversation into the right direction.

The panelists

Lucja Kowalewska, PhD is an Associate Professor and Head of the Department of Plant Anatomy and Cytology at the Faculty of Biology, University of Warsaw, where she leads the Plastid Nanomorphology Group. Her research focuses on the structural plasticity of plastid internal membrane networks, with a particular emphasis on cubic membrane configurations — including prolamellar bodies and gyrobodies — studied through an interdisciplinary combination of advanced electron microscopy, computational methods, and biophysical principles. She pioneered the application of mathematical tools from differential geometry and topology to plant cell biology, and developed the computational platforms SPIRE and GRANA for membrane structure analysis. She is Vice-Chair of the European Curvature and Biology Network (EuroCurvoBioNet, COST Action CA22153) and recipient of the 2025 National Science Centre Award in life sciences.

Jan Guzowski, PhD is a physicist specializing in experimental microfluidics and soft-matter and statistical physics. He has completed MSc in theoretical physics at University of Warsaw (2006) and PhD in theoretical and applied physics (2010) at Max Planck Institute for Metals Research (currently 'MPI for Intelligent Systems') in Stuttgart, Germany. During his postdoctoral years, he turned to experimental microfluidics and studied droplet self-assembly, e.g., in the so-called multiple emulsions at Institute of Physical Chemistry Polish Academy of Sciences in Warsaw, and their applications in tissue modeling and cell encapsulation at Princeton University, 2015-2017. After his return to IPC PAS Dr. Guzowski established his own research group focusing on soft granular matter and tissue engineering. His most recent research interests include new methods of droplet and particle manipulation for cell encapsulation and tissue engineering at the microscale, e.g. via the assembly of cell-microcarriers (particles and/or droplets) into mesostructures, for applications in basic developmental biology (e.g. angiogenesis) or modelling of cancer microenvironments in preclinical drug development. He is a co-founder of a spin off company Living Networks sp. z o. o.

Łukasz Andrzejewski, PhD is an expert in R&D strategy and digital product development. He graduated from the University of Wrocław and the University of Warsaw, and completed postdoctoral fellowships at The New School for Social Research and the University of Haifa. His professional experience includes roles in healthcare-focused venture capital funds and global technology companies. He currently leads the Discovery Department at Scope Fluidics, where he oversees the creation and development of special purpose vehicles in the deeptech, medtech, and life sciences sectors.

D-2: Science in Action: From Education to Media

In today's broad scientific landscape, it is not only discovery that matters, but also how new research is communicated and implemented. This creates a challenge for the academic community – to learn how to speak about and translate their scientific findings beyond the lecture hall.

The planned panel will provide a space where science communicators, reaching diverse communities and audiences, will discuss the challenges involved in encouraging people to engage with evidence-based, scientifically verified knowledge. The speakers will present different perspectives on connecting with online audiences through social media, introducing scientific solutions into the world of culture, and implementing sustainable practices within the film industry. They will also reflect on how both the scientific community and its surrounding networks approach the dissemination of knowledge.

The discussion will be moderated by **Zuzanna Opalska**, who will guide the conversation, highlight key questions, and foster an open exchange of ideas on the role of science in times of fake news and information chaos.

The panelists

Professor Katarzyna Siuzdak works at one of the institutes of the Polish Academy of Sciences, where she and her team focus, among others, on the fabrication of photoactive materials and electrochemical sensors. She studied biotechnology and technical physics simultaneously. In 2012, she earned her PhD in chemical technology, and 13 years later she was awarded the title of Professor in mechanical engineering. She is a recipient of the START scholarship from the Foundation for Polish Science (FNP), a scholarship from the Ministry of Science and Higher Education for outstanding young scientists, and the W. Nernst Award for Technical Achievement. She currently leads an international M-ERA.NET project. Her passion is science communication, particularly on Instagram, where through her profile @science_mission, she demonstrates how knowledge from the exact sciences is useful in everyday life, how to verify information found online, and how not to be misled by “the latest reports from American scientists”. In 2024, she was recognised as one of 12 outstanding female researchers from Gdańsk. Her outreach activities have been distinguished in the Science Populariser 2023 competition and the Silesian Science Festival (POP-science Award).

Professor Katarzyna Koziak is a Professor at Medical University of Warsaw, Head of the Department of Biochemistry and Nutrition, member of The Sectoral Skills Council for Life Sciences, led by BioForum. She conducts research at the intersection of biology, biochemistry, pharmacology and translational medicine, focusing, among others, on the mechanisms of inflammation and developing innovative therapeutic and health-promoting solutions. She is also a co-founder and Chief Scientific Officer at BioResearch Pharma, where she is responsible for the development of projects in the field of drug repositioning.

Marta Mielcarek is a cultural practitioner working at the intersection of culture, the creative industries, and social impact. She is the co-founder of the Slow Media Production Foundation, which promotes sustainable development in culture and media. She recently launched Slow Circle, an experimental podcast initiative exploring dialogue, civic reflection, and the role of culture and media in fast-changing, polarised times. A graduate of Journalism and of Theatre Production and Arts Organisation at the University of Łódź, she works on projects across the cultural and creative sectors. Her main interests include storytelling, cultural management and policy, artistic research, and sustainability.

W-1: Microbiology in Practice

*Aleksandra Wichrowska*¹

¹ Field Application Specialist, [Bacteromic](#)

This workshop will primarily focus on the essentials of antimicrobial susceptibility testing (AST) within the clinical laboratory. We will delve into the decision-making process behind when to perform a susceptibility test and explore various methodologies available. The session will cover a broad range of diagnostic techniques—from conventional to cutting-edge—and provide insights into the interpretation of AST results.

W-2: Science commercialisation - how to turn scientific ideas into real-world solutions

*Michał Jeska*¹

¹ [Medical Innovation Institute \(MII\)](#)

This hands-on workshop is designed for students, researchers, and early-stage innovators who want to turn scientific ideas into real-world solutions.

Biotechnology and medtech projects often start with strong scientific foundations, but the key challenge is translating them into products that solve real user and market needs. This session will guide participants through that transition - from concept to validation.

During the workshop, participants will learn how to:

- identify real problems faced by patients, clinicians, or industry stakeholders,
- formulate clear value hypotheses,
- test and validate their ideas in a structured and practical way.

Using real-life examples from biotech, medtech, and technology startups, we will demonstrate how to move beyond intuition and build solutions grounded in evidence and user insight. Participants will be introduced to practical tools such as Empathy Map, Lean Canvas, and Customer Journey Mapping, adapted to the context of life sciences and healthcare innovation.

This session is particularly valuable for those who want to bridge the gap between research and commercialization and start thinking about their projects not only as scientific achievements, but also as future products with real impact.

O-1: Hijacking Host Immunity: How *Fasciola hepatica* Challenges Vaccine Development

Alicja Laskowska¹, Mateusz Pękacz¹, Katarzyna Basała¹, Anna Zawistowska-Deniziak^{*,1}

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Parasitic helminths are masters of immune manipulation, capable of reshaping host responses. Among them, *Fasciola hepatica*, the causative agent of fasciolosis, poses a significant burden in both human health and livestock production worldwide. Despite considerable efforts, the development of an effective vaccine remains elusive.

Our previous observations suggests that *F. hepatica* molecules present in excretory-secretory products can suppress chemokine signals critical for antigen presentation and immune cell recruitment. Such interference could partly explain the limited success of vaccine candidates.

In this study, we sought to elucidate the immunomodulatory effects of *F. hepatica* excretory-secretory products (*Fh*ESPs), with a focus on their ability to alter chemokine responses in human dendritic cells and macrophages. *Fh*ESPs were obtained from adult parasites isolated from experimentally infected ovine livers and used to stimulate human immune cells *in vitro*. Chemokine and cytokine secretion, surface marker expression, and phagocytic activity were assessed.

Our results demonstrate that *Fh*ESPs modulate chemokine secretion in both dendritic cells and macrophages, while simultaneously reshaping their phenotypic and functional profiles. To further identify the components responsible for these effects, we compared whole *Fh*ESPs, EV-depleted *Fh*ESPs, and extracellular vesicles (EVs). Strikingly, the immunomodulatory effects were largely preserved in EV-depleted *Fh*ESPs, whereas EVs alone induced only limited changes, indicating that the key immunosuppressive factors reside predominantly in the soluble fraction of *Fh*ESPs.

Taken together, our findings support the concept that *F. hepatica* actively disrupts chemokine-mediated immune communication, potentially undermining the effectiveness of vaccine-induced responses. Unraveling these mechanisms provides critical insight into parasite-driven immune evasion.

Financial support provided by NCN, project nr: 2021/43/D/NZ6/01555.

Keywords: *Fasciola hepatica*, vaccine, immunomodulation, immune cells, chemokines

O-2: Parasite-derived extracellular vesicles regulate NLRP3 inflammasome signalling in an *in vitro* model of intestinal inflammation

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Extracellular vesicles (EVs) released by parasitic helminths have recently emerged as important mediators of host–parasite communication, particularly in the modulation of host immune responses. The NLRP3 inflammasome is a cytosolic multiprotein complex of the innate immune system that senses cellular stress and promotes the activation of caspase-1, leading to the secretion of pro-inflammatory cytokines such as IL-1 β and IL-18. *Anisakis simplex*, a parasitic nematode responsible for anisakiasis in humans, has been shown to secrete EVs that may influence inflammatory processes in the intestinal epithelium.

The aim of this study was to investigate the effect of *A. simplex* EVs on inflammatory responses associated with the NLRP3 inflammasome pathway, with particular emphasis on the gene expression and secretion of its downstream effector cytokines, IL-1 β and IL-18, together with caspase-1 activity, in an *in vitro* model of human intestinal epithelial cells (Caco-2). Cells were cultured under standard conditions and divided into four experimental groups: control cells, cells with cytokine-induced inflammation, cells treated with *A. simplex* EVs, and cells co-treated with cytokines and EVs. The gene expression and protein secretion was quantified in cells and culture supernatants using real-time PCR and enzyme-linked immunosorbent assays (ELISA), respectively.

In particular, treatment with EVs alters the gene expression and secretion of IL-1 β and IL-18 compared to inflammation-stimulated cells. These findings provide new insights into the role of parasite-derived EVs in shaping host intestinal inflammation and contribute to a better understanding of immune evasion strategies employed by helminths.

This research was funded by the Rector’s Student Grant (6th Edition) of the University of Warmia and Mazury in Olsztyn (5/SGRVI/2024) and as part of a project funded by the National Science Centre, Poland (2023/49/B/NZ6/02078).

Keywords: *Anisakis simplex*, extracellular vesicles, inflammasome, ELISA gene expression

O-3: Cross-species differences in dsRNA detection and translation shutdown among influenza virus hosts

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Influenza viruses circulate between birds and mammals, and crossing species barriers drives the emergence of novel, potentially virulent strains. A critical component of host innate immunity is the detection of viral double-stranded RNA (dsRNA) by specific intracellular sensors. The RIG-I-like receptor (RLR) pathway recognizes dsRNA to initiate signaling cascades that drive interferon expression, while the activation of PKR and RNase L pathways leads to the shutdown of cellular translation to restrict viral replication. This study aimed to compare these mechanisms of dsRNA recognition and intracellular translation arrest across three key species involved in the influenza virus evolution chain: human (A549 cell line), chicken (DF-1), and duck (CCL141). To mimic viral replication products, short and long dsRNA transcripts were designed and synthesized using *in vitro* transcription (IVT). Following transfection, translation inhibition was evaluated by assessing the phosphorylation levels of PKR and eIF2 α , alongside global translational activity measured by puromycin incorporation. Western Blot analysis was employed for protein detection, utilizing sodium arsenite as a positive control. Furthermore, the immunogenicity of these dsRNA variants was quantitatively assessed using a luciferase IFN- β reporter assay. Our findings indicate that chickens cannot sense short 5'-ppp dsRNA, while ducks and humans detect most dsRNA species, except short 5'-OH dsRNA. Additionally, long dsRNA strongly shuts down translation in human cells. These results provide crucial molecular insights into varying host susceptibilities to influenza and help elucidate the evolutionary divergences that allow ducks to act as asymptomatic viral reservoirs.

Keywords: dsRNA sensing, translation shutdown, viral RNA, innate immunity, cross-species differences

O-4: Proxyphylline Derivative versus Classical Calcineurin Inhibitors: A Novel approach for Controlling Cytokine Storms in SARS-CoV-2 Infection

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SARS-CoV-2 is a viral infection that involves a cytokine storm. A cytokine storm is characterized by elevated levels of IL-6, IL-1 β , and TNF- α responsible for severe COVID-19. A new drug, PB-WUT-01, was tested as a compound intended to block the pathway of cytokine synthesis by inhibiting calcineurin. This new drug was designed using *in silico* techniques. We compared the immunomodulatory potential of our drug with classical calcineurin inhibitors: tacrolimus (FK506) and cyclosporine A (CsA).

We tested the drug using an *in vitro* model that simulates the immunological microenvironment of the respiratory tract. Human mast cells (HMCs) and peripheral blood mononuclear cells (PBMCs) were exposed to three SARS-CoV-2 variants of concern: Alpha, Delta, and Omicron. SARS-CoV-2 induced a strong pro-inflammatory response, particularly involving increased secretion of IL-1 α , IL-6, and MCP-1. PB-WUT-01 significantly lowered the expression of key inflammatory mediators, including IL-6, TNF- α , and IL-17F, in both HMCs and PBMCs. This compound also modulated the expression of the ACE2 receptor, which may suggest a potential impact on the mechanisms of viral entry into cells.

Compared to classical calcineurin inhibitors, PB-WUT-01 exhibited more selective immunomodulatory properties—it effectively inhibited excessive cytokine production while preserving basic immune system activity. In contrast, FK506 showed a weaker suppressive effect under some of the analyzed conditions.

The structural features of PB-WUT-01 enabled improved interaction with the active site of calcineurin, potentially enhancing its ability to regulate the signaling pathway. Therefore, the developed compound represents a promising therapeutic candidate for controlling excessive inflammatory responses associated with viral infections.

These results suggest PB-WUT-01 may represent a next-generation calcineurin inhibitor, capable of modulating the immune response during SARS-CoV-2 infection.

Keywords: SARS-CoV-2, HMC, PBMC, Cyclosporine A, Tacrolimus, Proxyphylline

O-5: Toward a Genetically Modifiable Neutrophil Model: Functional Assessment of CEBP α -Induced Differentiation in K562 Cells

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Introduction

Neutrophils are a crucial component of the immune system, integrating antimicrobial and immunomodulatory functions through various strategies, including phagocytosis, generation of reactive oxygen species (ROS), and neutrophil extracellular traps (NETs). Owing to their complexity, neutrophils emerged as a major focus of biomedical research. However, neutrophil studies face significant challenges, including terminal differentiation, tendency to spontaneous activation, and resistance to genetic modifications.

To address this limitation, we developed a neutrophil-like cellular model using the K562 cell line, with inducible overexpression of the CEBP α , a major granulopoietic transcription factor. The CEBP α encoding sequence was introduced to K562 cells via a doxycycline-inducible Tet-ON lentiviral system.

Aim

Our research aims to assess the capacity of our model to perform neutrophil effector functions.

Methods

Phagocytosis and ROS generation were analyzed by flow cytometry. Differentiated cells were stained with a fluorochrome-conjugated CD11b antibody and exposed to FITC-labeled *E. coli* bioparticles (phagocytosis) or dihydrorhodamine 123 (ROS). For NET formation analysis, CD11b-positive cells were isolated by immunomagnetic separation, stimulated for 3 hours with PMA, PAF, LPS, or calcium ionophore, and stained with Sytox Green. NETs formation was analysed by fluorescence microscopy.

Results

Our findings indicate that our model is unable to perform phagocytosis. Both differentiated and undifferentiated cells were capable of generating ROS, although to a lesser extent than peripheral blood neutrophils. However, microscopic analysis revealed that differentiated K562-CEBP α cells can release NETs upon stimulation.

Conclusion

The obtained results indicate that our model mimics certain effector functions of neutrophils, yet further optimization is required to improve its functional efficiency.

Keywords: neutrophil-like cells; neutrophil; K562; CEBP α ; NETs

O-6: Human Induced Pluripotent Stem Cell-Derived Cardiomyocytes and Cardiac Progenitor Cells for Cell-Based Therapy in Murine Model of Myocardial Infarction

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Myocardial infarction (MI) is a leading global cause of death and results from sudden occlusion of coronary blood flow, leading to massive cardiomyocyte loss. Current therapies focus on reducing ischemia duration and limiting infarct size but do not restore damaged myocardium, which is replaced by non-contractile scar tissue.

Cell-based therapy has emerged as a promising approach for cardiac regeneration, however, the clinical efficacy of such therapies is limited. We aim to investigate the therapeutic potential of human induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CMs) and cardiac progenitor cells (hiPSC-CPCs) and hypothesize that hiPSC-CMs and hiPSC-CPCs may differ in engraftment efficiency due to their distinct proliferative and differentiation potential.

hiPSC-CPCs and hiPSC-CMs were obtained by differentiation of genetically modified hiPSC cell line, with GFP and luciferase expression, enabling detection of transplanted cells by fluorescence and bioluminescence. MI was induced by left anterior descending artery ligation in NOD-SCID mice, followed by immediate cell transplantation. The animals were observed and investigated for 12 weeks and then euthanized for histological assessment.

Our *in vitro* phenotypic characterization by flow cytometry showed that 35% of hiPSC-CPCs were OCT3/4⁺, while hiPSC-CMs lacked OCT3/4 expression. Differentiation from CPCs to CMs was associated with decreased GATA4 and increased cTnT expression. *In vivo*, hiPSC-CMs engrafted more efficiently than hiPSC-CPCs based on bioluminescence assessment. MI caused a significant decrease in cardiac functional parameters, which improved after cell therapy. Post-MI cardiac tissue showed no significant differences in myocardial fibrosis between groups.

Our results contribute to understanding the regenerative potential of hiPSC-derived cardiac cells for post-infarction repair.

This study was funded by SHENG-2 grant from the National Science Centre [2021/40/Q/NZ3/00165].

Keywords: Myocardial Infarction, Cell Therapy, Cardiac Regeneration, Human Induced Pluripotent Stem Cells (hiPSCs)

O-7: Bone Marrow Endothelial Cells as phagocytes: uptake of stressed erythrocytes and microparticles

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Bone marrow sinusoidal endothelial cells (BMSECs) play a key role in multiple processes and in maintaining the bone marrow microenvironment. Their primary function is to regulate the activity of hematopoietic stem cells and their differentiation into mature blood cells. BMSECs are also in close proximity to erythroblastic islands, where the final stages of erythropoiesis occur, suggesting a potential involvement of BMSECs in this process. Additionally, BMSECs express surface receptors that mediate the phagocytosis of damaged and stressed red blood cells (sRBCs), including Stabilin-2 (Stab-2) and Fc γ RIIb (FcRIIb).

In this study, we aimed to investigate the mechanisms of sRBC phagocytosis by BMSECs, with particular focus on the roles of Stab-2 and FcRIIb, as well as to assess the ability of BMSECs to internalize latex beads. For this purpose, we performed an in vivo approach using C57BL/6J mice. Stressed RBCs were generated by subjecting isolated erythrocytes to heat shock. In the first part of the study, mice were divided into groups depending on whether they received sRBCs or latex beads, and whether blocking antibodies against Stab-2 and FcRIIb were administered. Prior to injection, sRBCs were labeled with the fluorescent dye PKH26. In selected groups, mice were pre-treated with antibodies blocking Stab-2 and FcRIIb before administration of labeled sRBCs or beads. BMSECs were then labeled in vivo, and bones were harvested. Some of the bones were frozen and sectioned into thin slices for confocal microscopy analysis, while BMSECs were isolated from the remaining bones and analyzed by flow cytometry.

Preliminary results suggest that blocking Stab-2 and FcRIIb leads to increased phagocytosis of sRBCs, which may indicate the presence of alternative pathways mediating sRBC uptake by BMSECs. Moreover, BMSECs were shown to be capable of internalizing latex beads. Confocal analyses further demonstrated the presence of both sRBCs and latex beads near or within BMSECs.

Keywords: Bone marrow sinusoidal endothelial cells, red blood cells, latex beads, phagocytosis

O-8: Sf1 is translationally controlled through Igf2bp2 to regulate hematopoiesis

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RNA splicing consists of removal of introns and ligation of exons to form mature mRNA, expanding the coding capacity of the genome. This process is carried out by the spliceosome. One of the initial steps during splicing is the recognition of the branchpoint adenosine within introns, facilitated by binding of splicing factor 1 (Sf1). Defects in splicing are implicated in many diseases i.e. cancer. However, the role of splicing in homeostasis, particularly in stem cell differentiation, remains unclear.

Here, we analysed how the levels of Sf1 are regulated during activation of hematopoietic stem cells (HSCs). HSCs are a paradigmatic stem cell system with important translational value. We observed that Sf1 is dynamically regulated at the protein level during HSCs differentiation, in the absence of associated changes in mRNA, suggesting posttranscriptional mechanism of regulation. Strikingly, Sf1 is essential for the loss of stemness and differentiation. Therefore, our aim was to understand how Sf1 is regulated during blood differentiation.

To address this question, we analyzed the genomic locus of Sf1, revealing evolutionary conserved and highly structured 5' UTR. To assess the *cis*-acting factors, we generated the reporter system with Firefly luciferase controlled by deletion and substitution mutants of murine Sf1 5' UTR and measured luciferase activity. Regions crucial for efficient translation were identified (stem loops 2 and 3). Subsequently, to discover *trans*-acting factors, we performed RNA immunoprecipitation experiments with full-length or mutant, translationally deficient variant of Sf1 5' UTR. Coupled mass spectrometry analysis identified Igf2bp2, a well-studied 5' UTR binding translational activator, to be enriched in the full-length variant. We propose the new model of HSCs activation through upregulation of splicing factors translation, leading to spliceosome remodelling, resulting in global alternative splicing changes promoting differentiation.

Keywords: hematopoietic stem cells, RNA splicing, translation

O-9: Evolutionary pressure observed *in vitro* – a case of *Pseudomonas aeruginosa* bacteriophages developing lytic host adaptation

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Understanding interactions between bacteriophages and their bacterial hosts is essential for their application in the treatment of multidrug-resistant infections. *Pseudomonas aeruginosa* is an opportunistic pathogen that can cause severe infections of the lungs, wounds, and ears. Lytic phages are ubiquitous in the environment and play a key role in regulating bacterial populations and facilitating horizontal gene transfer. During co-infection, phages may exchange genetic material and exploit host molecular machinery, leading to the emergence of novel, potentially advantageous traits.

Based on these principles, Appelmans' protocol was applied. This method promotes genetic exchange between phages within a cocktail, aiming to expand their host range. In an experiment consisting of 10 rounds of the protocol, we investigated the evolution of lytic activity against *P. aeruginosa*. The experimental system was based on a controlled and limited genetic pool comprising 3 phages and 6 bacterial strains with varying initial susceptibility, including clinical isolates from cystic fibrosis patients. The resulting phage cocktail exhibited lytic activity against all tested strains, as well as additional ones from the collection. Five phage strains responsible for the broadened lytic spectrum were isolated.

Genomic analyses revealed strong adaptive dynamics shaping phage genomes. Using Sanger and next-generation sequencing, the isolates were assigned to the genera *Litunavirus* and *Luzseptimavirus*. Recombination analysis demonstrated that genomic regions encoding tail fiber proteins were particularly prone to variation. These proteins are critical for host recognition, receptor binding, and infection initiation, suggesting that recombination in these regions plays a key role in host range expansion. These findings highlight the potential of directed phage evolution in developing biotechnological tools for the treatment of severe bacterial infections and as an alternative to antibiotics.

Keywords: bacteriophages, recombination, *Pseudomonas aeruginosa*, host spectrum, molecular interaction

O-10: Microfluidic system for long-term imaging of optically thin bacterial biofilms to correlate phenotypic heterogeneity with local microenvironment

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Biofilm is a complex community formed by microorganisms as a result of their adhesion to surface and to neighboring cells. Bacterial adaptation to environmental conditions is supported by the biofilm mechanical characteristics and multiple levels of biodiversity, including phenotypic heterogeneity – a phenomenon, when population, in which cells share the same genetic information, shows differences in cell behavior. While the phenotypic heterogeneity has been confirmed in single bacterial cells, bulk cultures, and early stages of bacterial biofilm development (up to 48 h), this remains elusive for a mature biofilm due to methodology limitations.

Here, we demonstrate a method for a reproducible, long-term (> two weeks) *Escherichia coli* biofilm cultivation. We use a microfluidic system that consists of hundreds of microwells dedicated for growth of optically-thin biofilms, and parallel microchannels providing nutrients. This experimental setup is crucial for the characterization of parameters such as biofilm growth rate, growth depth, and gene expression in a mature biofilm under well-defined conditions. Only then, we could move to investigating quantitative, spatio-temporal distribution of selected phenotypes related with biofilm formation. We integrate GFP protein with transcription of genes associated with adhesion (*fimH*) and matrix production (*csdG*), known to exhibit heterogeneous expression. Alongside, we measure oxygen concentration and pH using fluorescent indicators integrated with microparticles. Using time-lapse microscopy and semi-automated image analysis, we extract spatially resolved fluorescently-labelled subpopulations within the biofilm. This approach may allow us to directly link local conditions to phenotypes diversification.

Our platform provides a quantitative framework for studying biofilms beyond early stages. It helps to understand non-genetic sources of variability in biofilm growth and may contribute to strategies of controlling biofilms.

Keywords: microfluidics, biofilm, bacteria, phenotypic heterogeneity, quantitative microbiology

O-11: The structural and protective role of myxoxanthophyll in stabilizing the photosynthetic apparatus of cyanobacteria

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Myxoxanthophyll is a unique, amphipathic carotenoid glycoside found exclusively in cyanobacteria. Due to its distinct structure, it plays a crucial role both in protecting cells from photooxidation and in the proper organization and stabilization of membranes. Despite its prevalence, the exact impact of myxoxanthophyll on cell physiology under abiotic stress conditions still requires detailed investigation.

The aim of this study was to determine the structural and protective role of myxoxanthophyll in the photosynthetic apparatus of the cyanobacterium *Synechocystis sp.* PCC 6803 under abiotic stress conditions. To achieve this, wild-type (WT) strain and the lacking myxoxanthophyll $\Delta cruF$ mutant were cultivated under control conditions and subjected to light stress as well as temperature stress.

Comparative analyses revealed that the lack of myxoxanthophyll alters the pigmentation profiles of the cells, resulting in an increased phycobilisome-to-chlorophyll ratio. These phenotypic shifts are accompanied by profound changes in membrane architecture and altered membrane fluidity, as assessed via electron microscopy and EPR spectroscopy. Interestingly, the mutant exhibits a protective compensatory mechanism by altering its carotenoid composition and accumulating zeaxanthin. Furthermore, while the photosystem I to photosystem II ratio remains unchanged in the mutant, high-light challenges reveal that the lack of myxoxanthophyll leads to a disruption in energy transfer from the phycobilisomes (PBS) to the photosystems.

These results show that myxoxanthophyll is not merely a passive photoprotectant, but a critical structural scaffold required to maintain the functional coupling of the light-harvesting antennae and to prevent the physical collapse of the thylakoid membrane system under environmental stress.

This work was part of the Student Research Projects program 2025/2026 edition at Faculty of Biophysics, Biochemistry and Biotechnology, Jagiellonian University

Keywords: cyanobacteria, *Synechocystis*, carotenoids, myxoxanthophyll, oxidative stress, thylakoids

O-12: Susceptibility of environmental *Pseudomonas isolates* to P2D1 tailocins reveals potential impacts on microbial populations beyond Soft Rot Pectobacteriaceae

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Tailocins are contractile, phage tail-like bacteriocins produced by many bacterial species. P2D1 is a tailocin originally characterized in the soft-rot pathogen *Dickeya dadantii* 3937, a plant pathogen belonging to the Soft Rot Pectobacteriaceae (SRP). This tailocin is encoded within a 22kb prophage-like genomic region most homologous to the genome of the Peduovirus P2 (hence P2D1: phage P2-like dickeyocin 1). Tailocins recognize susceptible bacterial cells and induce phage-like death by puncturing the cell envelope. Although their release requires lysis of the producer cell, tailocins are thought to enhance bacterial fitness under appropriate conditions by mediating interference competition.

Tailocins are commonly regarded as kin-restricted weapons targeting closely related strains. However, increasing evidence suggests that some may act across taxonomic boundaries. In this study, the target range of P2D1 was assessed against bacteria isolated from diverse soil- and rhizosphere-associated habitats. A total of 480 environmental isolates collected at three different locations were screened for susceptibility to P2D1 using a spot assay. Nine susceptible strains were identified and characterized phenotypically and genetically. Antagonism was confirmed to be mediated by P2D1 using a tailocin-deficient *D. dadantii* mutant.

Taxonomic identification revealed that the susceptible strains did not belong to the SRP group but instead represented three *Pseudomonas* species: *P. germanica*, *P. tensinigenes*, and *P. parakoreensis*. Plant tissue assays suggested contrasting ecological roles: some exhibited opportunistic pathogenicity and caused soft-rot symptoms, similar to those of SRP pathogens, whereas others reduced *D. dadantii*-induced soft rot, indicating potential relevance for biocontrol. Together, these findings support a broader ecological role for P2D1 beyond closely related taxa, suggesting that tailocins may influence microbial community assembly across taxonomic boundaries.

Keywords: Tailocin, *Dickeya dadantii*, Phage tail-like bacteriocin, *Pseudomonas*, Rhizosphere

O-13: Synthesis of gold nanoparticles modified with a dinucleotide cap analog as potential biomedical carrier

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The dynamic development of nucleic acid-based therapies, such as mRNA vaccines and gene therapies, requires effective methods for the stabilization and delivery of genetic material into cells. A key inspiration for new solutions is the natural 5' cap structure (m⁷GpppG), which protects mRNA and regulates the translation process. Synthetic cap analogs allow for controlled modulation of these functions, making them promising therapeutic tools.

The aim of this study was to develop a carrier combining the activity of cap analogs with the properties of nanomaterials. The research involved the synthesis of a cap analog modified at the N2 position, the synthesis of gold nanoparticles (AuNPs), and attempts to obtain an AuNPCap conjugate. Monodisperse gold nanoparticles with a diameter of approximately 5 nm were successfully synthesized. Due to solubility issues, the final target cap analog was not obtained; however, its nucleotide precursor containing a thiol linker (-SH) was successfully produced. To verify the concept, a model ligand exchange was performed using a sulfur-containing analog, confirming the formation of sulfur-gold bondson the nanoparticle surface. The developed AuNP functionalization method enabled the production of stable conjugates with controlled physicochemical properties.

The results indicate that the presence of cap analogs does not destabilize the gold nanoparticles, confirming the validity of the proposed concept. This work serves as a starting point for further optimization of the synthesis and the development of nanocarriers for nucleic acid-based therapies.

Keywords: 5' cap; Cap analogs; Gold nanoparticles (AuNPs); Eukaryotic translation initiation factor 4E (eIF4E)

O-14: Adapting an IMC foundation model for prediction of clinical features

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Imaging Mass Cytometry (IMC) provides spatially resolved measurements of over 40 protein markers in a single tissue section, enabling detailed characterization of tissue architecture and the tumor immune microenvironment. As public IMC resources increasingly include clinical annotations, an important question is whether representations learned by large pretrained models can be transferred to clinically relevant prediction tasks.

We address this question using ImmuVis, a recently introduced foundation model for IMC trained by self-supervised marker reconstruction on 28 cohorts, 24,405 images and 265 markers. We adapt the model for clinical classification by replacing the reconstruction decoder with a hierarchical multiple-instance learning head that aggregates patch-level information into sample-level predictions. We then compare the predictive utility of frozen pretrained embeddings against task-specific fine-tuning on data from 6 IMC panels obtained from 1659 patients from 5 cohorts.

Beyond predictive performance, we investigate interpretability by identifying molecular markers and spatial regions that contribute most strongly to classification decisions, and by assessing whether these patterns are consistent with current biological and medical knowledge. Our study evaluates foundation modeling as a practical starting point for clinically oriented analysis of IMC cohorts and explores when additional fine-tuning is needed for downstream use.

Keywords: Imaging Mass Cytometry; deep learning; foundational models; clinical prediction; model interpretability

O-15: $\text{Fe}_3\text{O}_4@AuNPs@Au@Pt$ nanozymes as promising multifunctional agents for biomedical diagnostic and anticancer applications

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This work presents the rational design and synthesis of multifunctional hybrid magnetic nanoparticles based on iron(II,III) oxide (Fe_3O_4), engineered as $\text{Fe}_3\text{O}_4@AuNPs@Au@Pt$ nanostructures. The developed system integrates magnetic, plasmonic, and catalytic components within a single platform, enabling dual functionality in anticancer therapy and biosensing applications.

The obtained nanostructures were analyzed using Vis–NIR spectroscopy, ζ -potential measurements, electron microscopy (SEM, TEM), elemental analysis (SEM-EDX, EDXRF), and vibrating sample magnetometry (VSM), confirming the formation of core–shell architectures with controlled surface composition. The nanostructures exhibited enhanced absorption in the near-infrared region (808 nm), within the biological transparency window, demonstrating strong potential for photothermal therapy (PTT) through efficient light-to-heat conversion.

The catalytic activity of $\text{Fe}_3\text{O}_4@AuNPs@Au@Pt$ was evaluated using peroxidase-like substrate (TMB), revealing high catalytic efficiency and the ability to generate reactive oxygen species (ROS), including hydroxyl radicals. This catalytic behavior is critical not only for chemodynamic therapy (CDT), but also for signal amplification in immunoassays such as ELISA and ELASA. Furthermore, surface functionalization with aptamers and antibodies enables selective recognition of cancer biomarkers, positioning these nanostructures as versatile diagnostic labels.

In vitro studies on normal (HaCaT) and cancer (A375) skin cell lines demonstrated selective cytotoxicity toward cancer cells, while maintaining low toxicity toward healthy cells, confirming their biocompatibility and therapeutic relevance.

$\text{Fe}_3\text{O}_4@AuNPs@Au@Pt$ nanozymes represent a promising theranostic platform that combines targeted cancer detection with synergistic PTT/CDT treatment, offering potential for advanced biomedical applications.

Keywords: nanoparticles, photothermal therapy, chemodynamic therapy, diagnostic

O-16: Reductase-mimicking nanozymes for novel signal generation strategy in paper-based immunoassays

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Recent advances in paper-based diagnostic systems have significantly improved infectious disease detection, as demonstrated during the COVID-19 pandemic. However, conventional lateral flow assays (LFAs) still exhibit limited sensitivity and relatively high detection thresholds, which may lead to false-negative results and reduced diagnostic reliability.

To address these limitations, nanomaterials with enzyme-like activity (nanozymes) have emerged as promising alternatives to natural enzymes due to their superior stability under varying environmental conditions. In this study, we investigate the use of nanozymes to enhance LFA sensitivity and lower detection limits. Unlike most existing approaches based on peroxidase-like activity (e.g., TMB oxidation), which require unstable oxidizing agents such as H₂O₂ and are prone to non-specific reactions, we focus on nanozymes catalyzing reduction reactions.

Bimetallic nanoparticles were synthesized via chemical reduction. Transition metal precursors, including Na₂PdCl₄, H₂PtCl₆, and HAuCl₄, were combined in various molar ratios to produce nanostructures exhibiting catalytic activity toward the reduction of tetrazolium salts, resazurin, and nitrophenol, which were repurposed as novel substrates for signal generation in point-of-care devices. The nanoparticles were characterized using DLS, UV-Vis spectroscopy, and TEM. Selected nanozymes were conjugated with antibodies targeting biomarkers such as C-reactive protein and ferritin, and incorporated into LFA systems as catalytic labels for signal amplification.

The developed nanozyme-based LFAs demonstrated significantly improved sensitivity, enabling detection of biomarkers at lower concentrations than conventional gold nanoparticle assays. This approach expands the capabilities of paper-based diagnostics and supports the development of more sensitive, reliable point-of-care devices.

Keywords: Nanozymes, Point-of-Care, LFAs, reductase-like activity

O-17: Purification of human poli(A) polymerase using ÄKTA liquid chromatography system

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The expression of protein-coding genes in eukaryotic cells is a multi-step process. First, transcription occurs with the involvement of RNA polymerase II. Next, the resulting precursor mRNA (pre-mRNA) undergoes maturation through processes such as capping at the 5' end, splicing, and polyadenylation at the 3' end. The final stage involves endonucleolytic cleavage of the pre-mRNA molecule and the addition of polyadenosine tracts, about 200 nucleotides long. Such polyadenosine tails are in almost all eukaryotic mRNAs. Additionally, they are essential for the transport of mRNA from the nucleus to the cytoplasm, translation, and protection of mRNA from degradation. The process of polyadenylation is mediated by several proteins: the cleavage and polyadenylation specificity factor (CPSF), the cleavage stimulation factor (CstF), cleavage factors I (CFI) and II (CFII), the 3' end maturation factor (PABPN1), and poly(A) polymerase. The latter enzyme is responsible for adding adenine residues to the 3' end after cleaving the pre-mRNA.

The aim of this study was to purify poly(A) polymerase in order to use this protein to reconstitute the polyadenylation reaction under *in vitro* conditions. For this purpose, a bacterial expression system was employed to produce poly(A) polymerase, followed by a multi-step purification procedure using liquid chromatography with the ÄKTA system. As a result of this work, a promising purification protocol for poly(A) polymerase was developed, enabling the acquisition of protein of sufficient quality for further analyses. The obtained results provide a foundation for future studies on the *in vitro* reconstitution of the polyadenylation reaction and on elucidating its mechanism. The enzyme will also be used for the subsequent reconstruction of the cleavage and polyadenylation complex, which will support research focused on the interaction between RNA cleavage and transcription termination.

Keywords: *Porphyromonas gingivalis*, lipoprotein, export signal, protein secretion

O-18: Identification of new components of the lipoprotein secretion system in *Porphyromonas gingivalis*

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Porphyromonas gingivalis plays a significant role in the development of periodontitis, a chronic inflammatory disease of the gums. Surface lipoproteins (SLPs) are among the key virulence factors of this pathogen. Important group of SLPs are proteins forming fimbriae – proteinaceous filaments that enable bacterial adhesion to host cells and coaggregation with other bacteria in oral microbiota. Although they are essential for the pathogenesis of *P. gingivalis*, the molecular mechanism underlying their secretion remains poorly understood. It is proposed that four proteins – PgmA, FimX, PgmB and FimY, form a putative lipoprotein secretion system (LSS). However, due to the complexity of secretion systems, these components alone are not sufficient to accomplish all steps of this process. The main objective of this study is to identify and characterize proteins associated with the lipoprotein secretion system, in order to gain a better understanding of the molecular basis of this mechanism.

The experimental strategy involved the isolation of His-tagged LSS complexes using a membrane-permeable cross-linker (DTSSP). Subsequently, the gene encoding the interacting protein was deleted, and the resulting effects on the bacterial phenotype were evaluated. The findings indicate a potential association between the LSS and the Type IX secretion system. These results provide new insights into the functional role of the LSS and may contribute to the development of targeted therapeutic approaches.

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Keywords: *Porphyromonas gingivalis*; lipoproteins, secretion systems

O-19: “The LES(s) I know, the better” – characterisation of Lipoprotein Export Signal in the unique secretion system of *Porphyromonas gingivalis*

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Surface lipoproteins of Bacteroidetes perform crucial functions in bacterial physiology and in interactions with the host, yet the mechanisms that determine whether these proteins remain hidden within the cell envelope or become exposed on the cell surface have remained unresolved. In *Porphyromonas gingivalis*, a major oral pathogen, a dedicated lipoprotein secretion system has been identified, but the molecular signal that targets the lipoprotein to the outer membrane surface has not been fully defined. Sequence comparisons of known surface lipoproteins revealed a conserved region located immediately downstream of the cleaved signal peptide, consisting of an N-terminal cysteine followed by negatively charged residues. This motif was proposed to act as a Lipoprotein Export Signal (LES), potentially serving as a molecular tag that directs lipoproteins to the bacterial surface.

In this study, we characterized the sequences of the LES in *P. gingivalis*. Using homologous recombination in the bacterial genome, selected negatively charged residues within the export motif were mutated in order to determine their contribution to lipoprotein targeting. Our results demonstrated that alterations within this short N-terminal motif affected lipoprotein localization, supporting its key role in surface export.

Defining the LES not only helps explain how surface lipoproteins are selectively transported, but also opens new perspectives for studying host–pathogen interactions.

This study was supported by a Polish National Science Centre grant to MM (UMO-2023/51/D/NZ1/02675). We gratefully acknowledge Polish high-performance computing infrastructure PLGrid (HPC Center: ACK Cyfronet AGH) for providing computer facilities and support within computational grant no. PLG/2025/018356.

Keywords: body mass, gut microbiome, polycystic ovarian syndrome, vaginal microbiome

O-20: Unlocking non-canonical rescue of the ubiquitin-proteasome system under functional compromise

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Efficient protein degradation by the ubiquitin–proteasome system (UPS) is essential for cellular proteostasis. While acute proteasome stress can activate compensatory transcriptional responses, mechanisms that sustain UPS function during prolonged proteasome impairment remain poorly understood.

Using *Caenorhabditis elegans*, we investigated transcriptional regulators that influence UPS resilience under conditions of reduced proteasome activity. A targeted RNAi analysis identified the Mediator subunit MDT-15 as a factor modulating UPS performance under proteasome stress. Depletion of MDT-15 restored proteasome-dependent degradation in animals with compromised proteasome β -subunit, indicating activation of a compensatory pathway that maintains UPS function. This effect appears highly specific, as knockdown of other Mediator components did not reproduce the phenotype.

Depletion of the transcription factor SBP-1, a known functional partner of MDT-15 in lipid metabolic gene regulation, produced a similar phenotype, suggesting that these factors participate in a shared transcriptional program influencing UPS activity. In contrast, canonical regulators of stress and metabolic signaling, including HSF-1 and NHR-49, were dispensable for this response.

Together, our findings reveal a previously unrecognized transcriptional layer of proteasome regulation. Preliminary experiments in mammalian cells suggest that elements of this mechanism may be evolutionarily conserved.

Keywords: ubiquitin–proteasome system, transcriptional regulation, *Caenorhabditis elegans*, Mediator complex

O-21: Deep Learning in tumor immune microenvironment analysis from multiplex imaging

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In recent years, the tumor immune microenvironment (TIME) has been shown to correlate with the effectiveness of immunotherapy. However, mechanisms underlying this relationship remain unclear. In our research, we investigate the potential of deep learning methods applied to high-resolution multiplex spatial images capturing the molecular organization of TIME. Our goal is to extract biologically relevant features, learn informative representations, and develop machine learning tools that support the characterization of tumor structure within tissue samples. To this end, we study a multimodal setting combining multiplex immunofluorescence (mIF) imaging with expert-derived histopathological annotations of regions of interest. Because these data sources are not directly aligned, we employ and extend a registration approach designed to handle noise and improve correspondence between modalities. The resulting annotations enable supervised learning on the associated mIF images.

As a baseline, we use a U-Net architecture for image segmentation. Our objective is to automatically identify histopathologically relevant tumor regions and their boundaries, and subsequently classify subregions of these border areas into molecularly distinct clusters, for example reflecting differences in the local immune microenvironment. Preliminary experiments show promising performance in tumor-region detection, supporting the feasibility of this approach.

We are currently evaluating more advanced architectures and extending training to larger datasets that include additional tumor types. We also investigate whether models trained on tumor edge tiles can generalize to other tissue regions. We anticipate that this work will contribute to a better understanding of tumor organization and tumor-immune interactions.

Keywords: Tumor Immune Microenvironment, Multiplex Immunofluorescence, Machine Learning

O-22: High-Sensitivity Sequential cfDNA Testing of MDM2 and HSP90AB1 Copy Levels for Progression Monitoring in Soft Tissue Sarcoma Patients

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Soft tissue sarcomas (STS) are rare malignancies with high risk of recurrence and metastasis. Early identification of progression remains a clinical challenge. Liquid biopsy of circulating cell-free DNA (cfDNA) offers a real-time reflection of disease dynamics. We evaluated whether sequential assessment of *MDM2* and *HSP90AB1* copy levels in cfDNA improves early progression detection and disease monitoring in STS patients.

A cohort of 27 STS patients was prospectively monitored. Blood samples (Blood 1-6) were collected at predefined points from diagnosis to 6 months post-surgery. cfDNA was isolated, and *MDM2* and *HSP90AB1* copy levels were quantified across all points using ddPCR. To evaluate the sequential algorithm, we utilized *MDM2* levels after the first chemotherapy cycle (Blood 2) and *HSP90AB1* levels after radiotherapy (Blood 3). Differences between progression and progression-free groups were analyzed (Mann–Whitney U test), assessing individual and sequential performance.

MDM2 (Blood 2) and *HSP90AB1* (Blood 3) copy levels significantly differentiated the progression and progression-free cohorts ($p=0.018$ and $p=0.010$, respectively). Both individual tests demonstrated 100% sensitivity for detecting progression. However, when applied independently, *MDM2* and *HSP90AB1* yielded specificities of 71.4% and 76.2%, leading to high false-positive rates. Strikingly, the sequential diagnostic algorithm (*MDM2* in Blood 2 followed by *HSP90AB1* in Blood 3) improved overall specificity to 81.0% while perfectly maintaining 100% sensitivity.

Sequential evaluation of *MDM2* and *HSP90AB1* in cfDNA is a promising, minimally invasive strategy for monitoring STS dynamics. This orthogonal validation approach successfully reduces false-positive classifications (improving specificity) without compromising the detection of true progressors (100% sensitivity), highlighting its utility for precise clinical surveillance.

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Keywords: soft tissue sarcoma, liquid biopsy, cfDNA, sequential testing, disease monitoring

O-23: *In vitro* studies on the anticancer properties of graphene oxide aerosol, using pancreatic ductal adenocarcinoma cells as a biological model

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Due to its unique properties, graphene oxide (GO) can be considered a potential candidate for cancer therapies. Because of its specific structure of its sharp-edged flakes, it is capable of mechanically damaging cancer cells, leading to their death. Because this nanomaterial doesn't form a uniformly flat surface, GO doesn't provide a surface conducive to adhesion, which is crucial for cell attachment to the extracellular matrix (ECM). Additionally, GO influences molecular pathways in cancer cells. Given the properties of GO, it was decided to explore this issue through research on a biological model.

The study utilized a GO aerosol produced using the Bag-On-Valve (BOV) method, which allows for the application of this nanomaterial via surface spraying, making it easy to apply. The biological models used were pancreatic ductal adenocarcinoma (PDAC) cells from the BxPC-3 and AsPC-1 lines.

The study included a physicochemical analysis of a GO aerosol with a concentration of 4.5 g/L, including an assessment of particle stability and size using zeta potential and DLS analysis, as well as an analysis of chemical composition using spectroscopic methods (LIBS, EDX, ATR-FTIR). Scanning transmission electron microscopy (STEM) was used to show the ultrastructure of the GO surface.

In vitro studies of the effect of GO aerosol on cancer cells were based on morphological analysis using scanning electron microscopy (SEM), assessment of metabolic activity via the XTT assay, evaluation of migration using a wound -healing assay, and assessment of gene expression involved in epithelial-mesenchymal transition (EMT) using RT-qPCR.

The results obtained indicate changes in the morphological and functional state of cancer cells under the influence of GO. The observed morphological changes, reduced viability, and alterations in the expression of genes involved in EMT offer great promise for the future, and the analyses conducted can serve as a basis for further research on this topic.

Keywords: BxPC-3, AsPC-3, pancreatic adenocarcinoma, GO, graphene oxide

O-24: Microfluidic-Assisted Precise Bioprinting of Hydrogel Micro-Bioreactors

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Current drug screening methodologies primarily rely on standard 96-well plates, which limits assay throughput and necessitates costly automated systems. Microfluidic technologies have emerged as an alternative, enabling the production of hydrogel microspheres that function as independent bioreactors for High-Throughput Screening. Dense hydrogel microgranule suspensions are excellent scaffolds for tissue engineering due to their porous structure, which facilitates critical oxygen and nutrient perfusion. However, their utility as granular bio-inks in precision bioprinting is significantly restricted by the inability to precisely deposit individual granules. Overcoming this challenge is the central motivation of my PhD project.

In my project, I focus on mastering single-bead deposition, which is essential for fabricating reproducible arrays of miniature tumor models. For microsphere production, I used methacrylated gelatin (GelMA), generating them in microfluidic devices using in-flow crosslinking under near-UV light. I investigated the flow of these dense suspensions using microfluidic systems simulating a printing nozzle geometry. My research diagnosed that flow irregularities and clogging are mainly driven by particle-particle and particle-wall interactions. The mechanisms of blockage include sieving, aggregation, and arch formation. Unlike hard particles, soft microgranules can deform to pass through constrictions, but the formation of granular aggregates is strongly attributed to the material's surface properties.

In conclusion, my results indicate that mitigating particle interactions is fundamental to preventing jamming. Further optimization of surface properties, which can be improved by optimizing the crosslinking process, is required to resolve these flow instabilities. Establishing spatial control at the level of individual granules will unlock the creation of biological assays based on precision-printed microscopic beads.

Keywords: Precise bioprinting, Granular bio-inks High-Throughput Screening, Microfluidics

O-25: Enzymes of microsomal fractions of *Arabidopsis* leaves, involved in the synthesis of esters of free fatty acids with primary alcohols – biochemical characterisation and substrates specificity

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Fatty acid ethyl esters and wax esters are metabolites found in animals, plants, and microorganisms. In plants, the primary functional role of wax esters (WEs) is related to their presence in cuticular waxes, a protective hydrophobic layer on the surface of leaves that prevents water loss and protects the plant against environmental stressors, such as insects or UV light. While much less is known about the roles of fatty acid ethyl esters (FAEEs) in plants, they have been found in pollen, fruit arils, as well as oils and leaves of certain species. It has been reported that enzymes utilizing free fatty acid for the synthesis of WEs and FAEEs are present in *Arabidopsis* leaves and wheat roots. The activity of these enzymes in wheat roots was found to be strongly stimulated by treatment with alloxymid (a grass herbicide), suggesting their role in stress response. However, the biochemical properties of these enzymes have not been characterized so far, which is the subject of the present study.

It has also been reported that free fatty acids accumulate in plant membranes, when plants are subjected to environmental stresses. These free fatty acids can lead to several biophysical changes in membrane structure and to increased lipid peroxidation. Consequently, an excess of free fatty acids can be very harmful for the plants. Therefore, the enzymes characterized in this study may be involved in protecting plant membranes from the harmful effects of free fatty acids by converting them into ester forms, which are not hazardous to the plant.

In the present research, we tested the effect of factors such as reaction time, temperature, pH and the presence of divalent ions on the activity of these enzymes. We also examined their substrate specificity towards different alcohols and free fatty acids. Additionally, the effect of tetrahydrolipstatin (a known lipase inhibitor) on the formation of both WEs and FAEEs was investigated.

Keywords: *Arabidopsis*, wax esters, fatty acid ethyl esters, free fatty acids

O-26: Fabrication of Laser-Induced Graphene Electrodes for Lactate Biosensing

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Sepsis is a life-threatening condition requiring rapid diagnosis, with blood lactate as a key biomarker of severity. In this study, laser-induced graphene (LIG) electrodes were fabricated via vector-mode laser processing (sheet resistance < 15 Ω/sq) and functionalized with lactate oxidase (LOx) and FMN-dependent lactate dehydrogenase (LDH) for enzymatic lactate biosensing. Electrochemical performance was evaluated by cyclic voltammetry in PBS, lactate, and redox-active solutions.

LDH-based biosensors showed stronger responses than LOx systems without requiring an external mediator, likely due to direct electron transfer (DET) between LDH and the LIG surface. This effect is attributed to intrinsic surface –COOH groups formed during vector-mode processing, enabling enzyme immobilization via EDC/NHS coupling and promoting efficient electron transfer. In contrast, LOx relies on mediator-assisted transfer, resulting in weaker signals.

In PBS with and without lactate, LDH-functionalized electrodes exhibited a 180.44% average increase in peak current, while LOx-based sensors showed a –12.30% response. In redox-mediated conditions, LDH sensors also outperformed LOx (58.37% vs. 40.81%), despite lower enzymatic activity (115 U/mL vs. 400 U/mL).

These results demonstrate that vector-mode LIG electrodes provide a rapid, cost-effective biosensing platform with inherent surface functionality, eliminating the need for additional linker steps. Future work will explore pyrene-PEG functionalization to further enhance enzyme immobilization and DET performance.

Keywords: Laser-Induced Graphene, Lactate Biosensor, Electrochemical Sensing, Direct Electron Transfer, LDH, LOx, Cyclic Voltammetry

O-27: Influence of Medium pH on *In vitro* Growth, Vegetative Propagation and Phytochemical Properties of *Drosera capensis* L.

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Drosera capensis L. is a carnivorous plant capable of nitrogen assimilation through specialized mechanism for trapping and digesting small organism. For many years, species from this group have attracted attention as ornamental plants and as a source of compound with potential pharmacological activity. Inappropriate substrate pH may adversely affect plant growth and morphology, which is well known to cultivators if the genera *Drosera*, *Sarracenia*, and *Dionaea*. It has also been shown that, in carnivorous plants, substrate pH may influence phytochemical composition and, consequently, the biological activity profile of plant extracts.

Seeds of *D. capensis* from the University of Warsaw Botanical Garden collection were sterilized and sown on sterile media containing half-strength Murashige and Skoog medium, 30 g/L sucrose and 2,5 g/L phytoigel, adjusted to pH 3.5, 4.2, 5.8.

Plants cultivated on media with lower pH showed better condition, higher vitality and more intensive biomass growth. Initial organoleptic assessment suggested an advantage of acidic media. These observations were confirmed by fresh mass weight measurements. The greatest increase was recorded for plants growth pH 3.5. In addition, extract obtained from plants cultured under the most favourable conditions were evaluated for antioxidant activity using the DPPH and ABTS assays, also their total phenolic content was determined. An additional problem in the *in vitro* culture of *D. capensis* at pH below 5.8 was limited vegetative propagation. To overcome this issue, a new medium composition was developed, enabling production of a greater amount of plant material and improving the usefulness of the culture system.

The results indicate that selecting the optimal medium pH is crucial for the growth *D. capensis* *in vitro* cultures and may also affect the biological properties and phytochemical quality of the obtained extracts, which is important from biotechnological and pharmacological perspectives.

Keywords: *Drosera capensis*, *in vitro* culture, substrate pH, phytochemical, pharmacy

O-28: Cytotoxic activity of geranyl resveratrol derivatives in cutaneous squamous cell carcinoma models

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Cutaneous squamous cell carcinoma (cSCC) is the second most common skin cancer, mainly caused by extensive UV exposure and immunosuppression. Standard treatment includes surgery, radiotherapy, and, in advanced cases, immunotherapy. Emerging strategies explore compounds such as resveratrol and its derivatives due to their unique properties.

Resveratrol is a plant-derived compound found in nuts, berries, and grapes, with high concentrations in grape skin and red wine. It possesses a broad spectrum of effects, including anti-inflammatory, antioxidant, anticancer, and cardioprotective properties. However, resveratrol alone is insufficient for cancer therapy, as its safe plasma concentrations do not reach levels required for effective cancer cell inhibition. There is significant interest in synthetic derivatives of this compound, aimed at improving its efficacy and bioavailability.

In this study, geranyl resveratrol derivatives (6F, 7F, 6E) were investigated, with resveratrol included as a reference compound. Cytotoxic activity was evaluated using a standard MTT assay to determine the half-maximal inhibitory concentration (IC₅₀). Furthermore, apoptosis induction was assessed using an Annexin V assay. The compounds were tested on two human SCC cell lines (SCL-II, CRL-1555) to evaluate their anticancer potential. In addition, the human fibroblast cell line BJ was included as a non-cancerous control to assess selectivity. The results reveal that compounds 6F and 7F exhibited the highest cytotoxic potential, with significantly lower IC₅₀ values compared to derivative 6E. In the SCL-II cell line, 6F displayed an IC₅₀ of 25.62 µg/mL, comparable to 7F (IC₅₀=25.13 µg/mL), while 6E showed higher IC₅₀ value of 101.15 µg/mL. Resveratrol displayed pronounced time-dependent activity, exhibiting a moderate cytotoxic effect after 24 hours of incubation and a notable decrease in cell viability after 48 hours. Further research is needed to identify more promising analogs of resveratrol.

Keywords: resveratrol derivatives, cutaneous squamous cell carcinoma, cytotoxicity; apoptosis, skin cancer

O-29: Impact of silver nanoparticles and ions on development and maturation of guppy (*Poecilia reticulata*)

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The aim of this study was to evaluate the effects of silver nanoparticles (AgNPs) and silver ions (Ag⁺) on the morphology, growth, and development of sex characteristics in guppy (*Poecilia reticulata*) from hatching over a 60-day period. Juvenile fish were exposed to AgNP concentrations of 0 (control), 0.01, 0.05, 0.1, and 1 ppm, as well as ⁺. At the end of the experiment, the fish were euthanized, measured, and weighed, and then processed for whole-body histological analysis. The highest mortality rates, reaching approximately 40%, were recorded in the 1 ppm AgNP and 0.01 ppm Ag⁺ groups, compared to about 10% in the control group. Fish exposed to 0.05, 0.1, and 1 ppm AgNPs exhibited the lowest proportion of individuals with developed secondary sexual characteristics, with over 70% of specimens remaining sexually undifferentiated. Additionally, these groups showed less developed oocytes and testes compared to the control. A similar pattern was observed in body weight and length. Fish in the 0.05, 0.1, and 1 ppm AgNP groups had the lowest body weights, with the minimum value recorded at 0.029 ± 0.023 g in the 0.05 ppm group. Interestingly, these individuals also displayed relatively greater body length, reaching 11.68 ± 3.31 mm, even exceeding the control group in this parameter. However, fish from the 0.05 ppm group had the shortest total length, measuring 12.59 ± 2.00 mm. All exposed groups exhibited a higher number of proliferating hepatocyte nuclei compared to the control group. Overall, the results suggest that exposure to AgNPs may delay sexual maturation, disrupt normal morphology and growth, and trigger detoxification responses in fish.

Keywords: silver nanoparticles, guppy, histology, ecotoxicology, silver ions

O-30: The presence of linezolid-resistant *E. faecium* and *E. faecalis* strains in wastewater treatment plant environments

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In the clinical setting, infections caused by vancomycin-resistant *Enterococcus spp.* (VRE), particularly *E. faecium* and *E. faecalis* strains, remain a significant problem. One of the key drugs used to treat infections caused by VRE is linezolid. Given its use, monitoring the presence of linezolid-resistant *Enterococcus spp.* (LRE) strains, both in clinical and non-clinical settings, may be helpful in understanding the mechanisms of antibiotic resistance and the transmission of resistant strains.

The aim of the study was to assess the prevalence of linezolid-resistant *Enterococcus faecium* and *Enterococcus faecalis* strains isolated from hospital and municipal wastewater samples and to detect selected virulence genes among resistant microorganisms.

Material for the study consisted of 510 wastewater samples. Bacteria were isolated using selective media for *Enterococcus spp.* and chromogenic media for the selection of LRE strains. The isolated strains were identified using the MALDI-TOF MS technique. The susceptibility to linezolid was assessed using the Sensititre ARIS HiQTM System (Thermo ScientificTM). The presence of selected virulence genes (*agg*, *ace*, *EfaAfs*, *gelE*, *pil*, *ebpA/B/C*) was assessed using a PCR.

Out of collected samples, 438 (86.7%) *E. faecium* and 67 (13.3%) *E. faecalis* strains were isolated. Most of the microorganisms were isolated from treated wastewater (n=148; 33.8%) and hospital sewage (n=93; 21.2%). The presence of at least one selected virulence genes was found in 28 isolated strains, for which minimal inhibitory concentration (MIC) of linezolid ≥ 8 . The *ebpC* gene was predominant among LRE strains.

These findings confirm the prevalence of linezolid-resistant *E. faecium* and *E. faecalis* strains in wastewater and highlight the need for continuous environmental surveillance to limit the spread of resistance for linezolid and other antibiotics.

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Keywords: *E. faecium*, *E. faecalis*, wastewater treatment plants, linezolid-resistant *Enterococcus spp.*, virulence genes

O-31: Is the Cure for Neuroblastoma Hidden in the Baltic Sea?

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Neuroblastoma (NBL), the most common solid tumor in children, remains one of the deadliest pediatric cancers. Up to 70% of patients diagnosed at advanced stages (III–IV) cannot be cured with currently available therapies, highlighting the urgent need for novel and more effective treatment strategies.

Cyanobacterial metabolites have emerged as a promising and underexplored source of potent anticancer compounds. This study aimed to identify bioactive metabolites against NBL derived from two cyanobacterial species: *Nostoc edaphicum* (CCNP 1411) and *Pseudanabaena cf. galeata* (CCNP 1313).

To obtain such bioactive compounds, the entire process was carried out, from the extraction of whole cyanobacterial metabolomes, through their subdivision into smaller groups of compounds, to the isolation and identification of individual metabolites with therapeutic potential. Fractions and isolated compounds were tested on two NBL cell lines (SH-SY5Y, IMR-32) and pediatric fibroblasts (HDFc) as a model of healthy cells. At each stage, cytotoxicity was evaluated using the MTT assay.

Fractions containing metabolomes of the cyanobacterial species tested showed 43.7–90.2% decrease in the viability of NBL cells. After subdivision of the effective fractions, some of them again reduced cell survival by 62.2–89.4%, some of which had no cytotoxic effect on healthy cells. Then single compounds were isolated and those with anticancer properties were identified. The next step is to preliminarily determine their mechanism of action.

These findings highlight the exceptional potential of cyanobacterial metabolites as new anticancer agents and position them as promising candidates for further development. Ongoing studies focus on identifying their molecular targets and evaluating their efficacy and safety in advanced 3D and then possibly *in vivo* models.

Keywords: Neuroblastoma, Cyanometabolites, Cancer research

O-32: HydroBioCell – Development of an innovative, biodegradable, and antimicrobial water filter based on bacterial cellulose

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Modern industry using industrial water in open and closed circuits often struggles with the problem of microbiological contamination of water systems. This can lead to the formation of a biofilm and slime on the installation, which results in a decrease in the efficiency of the processes and possible secondary contamination.

The aim of the research was to construct biodegradable water filters based on bacterial cellulose modified with chitosan with antimicrobial properties and to study the influence of process parameters on mechanical and functional properties. The key factors were: pulp mass and degree of hydration, freezing temperature and chitosan impregnation.

The selection of mass and hydration of the pulp resulted in the appropriate porosity. The temperature of -80°C eliminated the separation of water from the pulp. The direct addition of chitosan to the prepared cellulose determined the concentration needed in the composite to impart durable antimicrobial properties.

Prototypes were tested on a small scale using 200 ml filtration apparatus connected to a vacuum pump, through which bacterial suspension containing *Staphylococcus aureus* was filtered. To determine the effectiveness of pathogen removal and inactivation from water, droplet cultures were performed, analyzing the bacterial titre from samples before and after filtration.

Comparing filtration trials on composites containing pure cellulose versus the biopolymer with added chitosan, a significant decrease in the number of microorganisms on general growth media plates was observed compared to both the pure cellulose filter and the control sample prior to filtration. Additionally, a substantial improvement in filter strength was noted after the addition of chitosan due to increased resistance to tearing caused by the pressure of the flowing water.

The results suggest that the constructed prototypes can be used as an ecological industrial filter that effectively cleans and reduces microorganisms in water.

Keywords: bacterial cellulose, chitosan, antimicrobial composites, water filtration, biodegradability

P-1: Identification of Novel Natural Compound Derivatives Modulating Large-Conductance Calcium-Activated Potassium (BKCa) Channels

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Large-conductance calcium-activated potassium (BKCa) channels are widely expressed ion channels that regulate membrane potential and cellular excitability. By responding to both membrane depolarization and intracellular Ca^{2+} signals, BKCa channels participate in many physiological processes including vascular tone regulation, neuronal signaling, and potassium secretion. Due to these roles, BKCa channels are considered important pharmacological targets in the treatment of cardiovascular and neurological disorders. Their activity can be modulated not only by voltage and Ca^{2+} but also by small natural and synthetic molecules.

The study aimed to verify the hypothesis that the modulatory properties of chalcones toward the BKCa channel depend on their hydrophobicity. Chalcones are α,β -unsaturated ketones composed of two aromatic rings and represent a large family of natural compounds and their derivatives with diverse biological activities.

BKCa channel activity was analyzed using the patch-clamp technique, which enables electrophysiological recordings of single-channel currents. Experiments were performed on human astrocytoma U87 cells cultured under standard conditions. Selected natural and synthetic chalcone derivatives were applied during recordings to evaluate their effects on BKCa channel activity.

Preliminary results indicate that relatively more hydrophobic trans-chalcone and 4',5,7-tri-O-methyl-naringenin chalcone inhibit BKCa channel activity, whereas more hydrophilic butein and naringenin chalcone act as channel activators. These observations support the hypothesis that the modulatory effect of chalcones on BKCa channels is related to their polarity.

These findings suggest that chalcone derivatives may provide a useful basis for the future design of compounds targeting BKCa channels. Understanding how their physicochemical properties influence channel modulation may support the development of new pharmacologically active molecules.

Keywords: chalcones; potassium channel; patch-clamp technique; BKCa; astrocytoma

P-2: Modulation of the TRPA1 channel affects functional properties of human pancreatic stellate cells in a phenotype-dependent manner

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Human pancreatic stellate cells (hPSC) are key regulators of extracellular matrix (ECM) homeostasis and major contributors to fibrosis in chronic pancreatitis and pancreatic cancer. Under physiological conditions, hPSC remain quiescent, whereas inflammatory and profibrotic stimuli promote their activation, leading to cytoskeletal remodelling, altered motility and increased ECM production. Since the transient receptor potential ankyrin 1 (TRPA1) channel is a regulator of intracellular Ca²⁺ signalling, its modulation may affect these processes.

This study examined the effects of pharmacological TRPA1 modulation using the agonist ASP7663 and the antagonist A-967079 in three hPSC phenotypes: quiescent (qhPSC), spontaneously activated (shPSC), and TGF- β -activated (ahPSC). The analysis included cell migration, focal adhesion organisation, viability, proliferative activity (EdU incorporation, Ki-67) and intracellular ATP levels.

Inhibition/activation of TRPA1 had limited effects on cell death in qhPSC and shPSC, whereas higher concentrations of ASP7663 increased cell death in ahPSC despite lower TRPA1 expression. The three phenotypes also differed in migratory properties: qhPSC exhibited the highest migration speed and displacement, while ahPSC showed reduced motility and greater directional persistence. Consistently, ahPSC displayed a higher proportion of elongated focal adhesions than qhPSC. TRPA1 activation selectively impaired migration in qhPSC, without altering focal adhesion number or length. Notably, this reduction in migration was associated with decreased intracellular ATP levels, suggesting that altered cellular energetics may contribute to TRPA1-dependent regulation of hPSC motility.

These findings indicate that TRPA1 contributes to the regulation of hPSC (patho)physiology. By linking ion channel activity with cellular energetics and migration, TRPA1 may be a relevant target in pancreatic diseases associated with fibrosis.

Keywords: pancreas; hPSC; fibrosis; TRPA1; calcium signalling

P-3: Antifungal activity of a gemini surfactant against *Aspergillus flavus* isolated from feed

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Filamentous fungi contaminating feed pose a significant problem due to mycotoxin production and reduced raw material quality. Of particular importance is *Aspergillus flavus*, capable of synthesizing aflatoxins (mainly B1 and B2), which threaten human and animal health as well as food chain safety. Therefore, new substances with antimicrobial properties capable of limiting pathogen growth are being sought. A promising solution is the use of gemini surfactants – a new generation of cationic surfactants. These compounds possess two hydrophilic groups and two hydrophobic chains connected by a spacer, which gives them high surface activity and antifungal properties.

In this study, the antifungal activity of hexamethylene-1,6-bis(N,N-dimethyl-N-dodecylammonium) dibromide (12-6-12) was evaluated against a strain of *A. flavus* isolated from feed. The effectiveness of the compound was determined by measuring the minimal inhibitory concentration (MIC) and the minimal biocidal concentration (MBC). Additionally, mould growth dynamics were analyzed using the Biolog Odin system.

In samples containing 12-6-12 at the MIC concentration (0.466 mmol/L), significant inhibition of *A. flavus* growth was observed, whereas at the MBC concentration (0.931 mmol/L) no macroscopic growth was detected, indicating an inability to rebuild the mycelial structure. At lower concentrations, growth kinetics analysis showed significant prolongation of spore germination and a reduction in the overall growth rate. At $\frac{1}{2}$ MIC, the compound induced a spherical form of mycelial growth without sporulation, whereas at $\frac{1}{4}$ MIC it significantly disrupted sporulation but did not completely inhibit it.

The obtained results indicate that gemini surfactants are promising compounds capable of limiting *A. flavus* development, highlighting their potential in biotechnology and food safety.

Keywords: *Aspergillus flavus*; gemini surfactants; antifungal activity; feed contamination

P-4: Assessment of biofilm formation by *Escherichia coli* 8917 on common 3D-printing polymers: applications in biosensor and bioreactor design

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Bacterial adhesion followed by biofilm formation is a critical factor in the development of chronic and recurrent infections. Consequently, there is a growing demand for innovative approaches enabling rapid detection of microorganisms and effective monitoring of microbial growth dynamics in environmental samples. The present study aimed to evaluate commonly used 3D-printing polymers with regard to their susceptibility to bacterial adhesion and their potential application in the design of biosensors and bioreactors.

The dynamics of biofilm formation by uropathogenic *Escherichia coli* 8917 (UPEC) were assessed on selected polymeric surfaces, including acrylonitrile styrene acrylate (ASA), polyethylene terephthalate glycol (PETG), thermoplastic polyurethane (TPU), polyethylene with carbon black (PE+CB), polylactic acid with carbon black (PLA+CB), and polylactic acid (PLA). Bacterial counts were determined after 3, 6, 24, and 48 hours of incubation. Across all tested materials, a progressive increase in bacterial cell numbers was observed up to 24 hours, followed by a decline at 48 hours. This pattern suggests the establishment of a mature and structurally organized biofilm within 24 hours, independent of polymer type, and subsequent dispersion of bacterial cells into the surrounding environment.

Among the tested materials, ASA and PE+CB demonstrated the highest susceptibility to bacterial colonization, reaching 8.3 and 9.1 log CFU, respectively, after 24 hours. Although the remaining polymers exhibited comparatively lower bacterial loads, they still supported substantial bacterial adhesion. Overall, the results indicate that *E. coli* 8917 is capable of forming robust biofilms on all evaluated polymeric surfaces. Therefore, none of the proposed materials can be considered suitable as non-adhesive material for the fabrication of measurement chambers intended for microbial detection and growth monitoring in controlled systems.

Keywords: Biofilm formation; Uropathogenic *E. coli*; 3D-printing polymers; Biosensor design

P-5: Bridging the Gap: Optimizing Direct Reprogramming of Human Fibroblasts into Neurons for Morphological Analysis

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Traditional methods of generating human neurons, such as induced pluripotent stem cells (iPSCs), are limited by long differentiation timelines and the loss of age-related cellular features. Direct lineage reprogramming offers an alternative approach by bypassing the pluripotent stage, potentially preserving biologically relevant characteristics.

The aim of this study was to optimize a reproducible and time-efficient protocol for direct conversion of adult human fibroblasts into neurons, with a particular focus on generating cells suitable for quantitative morphological analysis.

Human primary fibroblasts were transduced with an inducible lentiviral all-in-one system encoding proneuronal transcription factors Ngn2 and Ascl1. Cells were cultured on Matrigel-coated glass coverslips and multiwell culture plates. A differentiation protocol lasting up to four weeks was implemented. Different media compositions and small-molecule supplements were tested to improve conversion efficiency and neuronal maturation. Neuronal identity was assessed using immunostaining and Western blot analysis of selected neuronal markers. To enable visualization and downstream quantification of neuronal morphology, cells were additionally transduced with a GFP construct under the synapsin promoter.

The study established an iterative framework for optimizing direct conversion conditions, including refinement of culture parameters and differentiation timeline. The current approach enables the generation of neuron-like cells within a shortened timeframe and supports preliminary morphological assessment, while further optimization is required to improve consistency and analytical robustness for quantitative dendritic analysis.

Keywords: Direct reprogramming, Human fibroblasts, Neuronal differentiation

P-6: Establishing a FingR-based system for imaging PSD-95 in NT2-derived neurons

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Dendritic spines are small protrusions that form the postsynaptic sites of excitatory synapses and are essential for synaptic plasticity. In schizophrenia, reduced dendritic spine density and impaired synaptic plasticity have been reported, and it is hypothesized that antipsychotic drugs may modulate these changes. These processes can be studied by visualizing key scaffolding proteins of the postsynaptic density, such as PSD-95. Traditional visualization methods have limitations: immunocytochemistry requires fixation and provides only static images, while PSD-95-GFP fusions may mislocalize and cause structural artifacts. To address these issues, FingR (*Fibronectin intrabodies generated with mRNA display*) technology was developed, enabling real-time imaging of endogenous proteins in living cells without affecting synaptic function or structure. The aim of this study was to establish a FingR-based system targeting PSD-95 in human NT2-derived neurons. Two genetic constructs were designed for genomic integration using the Sleeping Beauty system: a complex self-regulating PSD95.FingR-GFP construct and a simplified doxycycline-inducible version. Due to the large size and high GC content of the self-regulating FingR sequence, extensive cloning optimization was performed (e.g. Nested PCR, Gibson Assembly). Despite these efforts, assembly of the complex construct was unsuccessful; however, the simplified construct was successfully cloned and verified by sequencing. Furthermore, transfection protocols for NT2 cells were optimized, allowing us to initiate generation of a stable NT2 cell line expressing the FingR construct. These cells will be further differentiated into neurons to enable real-time monitoring of PSD-95 dynamics in response to antipsychotic treatment. In conclusion, this study highlights the technical challenges of engineering complex genetic tools and demonstrates the potential of FingR-based approaches for studying synaptic changes in NT2-derived neurons.

Keywords: FingR; molecular cloning; PSD-95; dendritic spines; NT2 cells

P-7: Evaluation of Cellular Stress-Related Proteins in the Cerebral Cortex of Trap1-Mutant Male Mice: A Novel Model of ASD

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Background: TNF Receptor Associated Protein 1 (TRAP1) is a mitochondrial-specific chaperone from the HSP90 family. It functions as an ATP-dependent dimer, ensuring the correct folding and stability of numerous mitochondrial substrate proteins. The mutation in TRAP1 gene p.Q639* was identified recently in two unrelated male ASD patients. In order to research the functional consequences of this specific TRAP1 variant, a knock-in mouse model with identical mutation that was identified in ASD patients was developed (*Trap1* p.Q641*). *Trap1* mutation resulted in decreased *Trap1* mRNA levels and absence of detectable *Trap1* protein levels in the brain of *Trap1* mutant mice.

Aim: This study aims to determine whether the mutation on *Trap1* p.Q641* affects the expression of other heat shock proteins (HSP) and mitochondrial protein *Nlr1* in the mouse cerebral cortex.

Methods: Cortical tissue was obtained from 8-week-old wild-type (WT) and *Trap1*-mutant mice. Protein expression was quantified via Western Blotting for a comprehensive panel including *Nlr1*, mitochondrial chaperones (*Grp75*), endoplasmic reticulum stress markers (*Grp78*, *Grp94*), and cytoplasmic chaperones (*Hsp27*, 70, 105). Tubulin and *Gapdh* were used as a loading control for densitometric normalization.

Results: Verification of the model confirmed the total absence of *Trap1* protein in *Trap1*-mutant mice. The quantitative assessment of mitochondrial, cytoplasmic, and ER-resident proteins provided a comprehensive overview of the cellular stress response machinery in the *Trap1* mutants. This analysis revealed differences in the levels of GRPs and HSPs in WT mice and those with a mutation in the *Trap1* coding gene. Additionally, *Trap1*-mutant mice exhibited lower levels of *Nlr1* in cortices, compared to wild-type controls.

Keywords: *Trap1*; Autism; Mitochondria; Cellular Stress; Mouse Model

P-8: Development and validation of a bioinformatic pipeline for Ion Torrent sequencing data in forensic SNP analysis

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Forensic DNA phenotyping (FDP) has emerged as a powerful approach for generating investigative leads in cases where conventional STR-based identification fails. By enabling the prediction of externally visible characteristics (EVCs) and biogeographical ancestry from trace DNA, FDP can support criminal investigations, disaster victim identification (DVI), and the analysis of human skeletal remains. Recent advances in massively parallel sequencing (MPS) technologies have further expanded the scope and accuracy of such predictions, while simultaneously increasing the demand for robust and automated bioinformatic solutions.

The aim of this study was to develop a comprehensive bioinformatic pipeline for the analysis and interpretation of MPS data generated using an Ion Torrent platform, and to apply this pipeline in the developmental validation of a custom SNP panel designed for simultaneous prediction of appearance traits and ancestry. The panel integrates 161 genetic markers associated with sex determination, pigmentation, hair morphology, and additional facial features, enabling extended phenotype inference beyond standard pigmentation traits.

The performance of the assay and the accompanying analytical workflow was evaluated according to forensic validation guidelines, including sensitivity, reproducibility, repeatability, inhibitor tolerance, species specificity, and the ability to analyse mixed, degraded, and low-template DNA samples, including bone-derived material. The results demonstrated high robustness and sensitivity of the method, with reliable genotyping achieved from low DNA input quantities. Importantly, the success of the analysis was primarily dependent on DNA quantity rather than degradation level, confirming the suitability of the approach for challenging forensic samples.

In conclusion, the combined MPS assay and bioinformatic workflow provides a sensitive, reliable, and practical tool for forensic DNA phenotyping.

Keywords: forensic DNA genotyping; bioinformatics pipeline; MPS sequencing; validation studies

P-9: Intracellular Complement Components Regulate Metabolism and Stress Response in Bone Marrow Stromal and Hematopoietic Cells

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Bone marrow stromal cells form a specialized microenvironment that provides both structural support and secretory signals, releasing numerous growth factors and cytokines that regulate hematopoietic stem cell (HSC) proliferation, differentiation, and survival. The intracellular complement system, particularly C3 and C5, is an integral part of innate immunity and modulates HSC function by influencing migration, adhesion, homing, and bone marrow colonization. Additionally, interactions between stromal cells and complement components are complex: stromal cells can regulate complement activation, while complement proteolysis products affect HSC signaling and adhesion within the niche, thereby shaping hematopoietic efficiency.

To investigate the underlying mechanisms of bone marrow stroma complosome, we analyzed stromal cells isolated from bone marrow and Lin- hematopoietic cells from WT, C3-KO and C5-KO mice. Using holotomography, RT-PCR and mitochondrial fluorescent markers, we evaluated cellular morphology, mitochondrial redox status and metabolic gene expression, including genes involved in mitochondrial respiration (ND2, CYTB, COXI, ATP6), glycolysis (HK, PFK-1, LDHA/B), the Krebs cycle (IDH1, CS), the pentose phosphate pathway (G6PD, SHPK) and β -oxidation (ACACB, CPT1C).

Our results indicate that stromal and Lin- hematopoietic cells exhibit different metabolic and mitochondrial characteristics. HSCs are more susceptible to metabolic disruptions and oxidative stress, with C3 or C5 deficiency leading to alterations in the expression of genes related to glycolysis and the PPP. In contrast, stromal cells maintain more stable metabolic activity, highlighting their supportive role within the hematopoietic niche. These findings imply that intracellular complement components are crucial regulators of mitochondrial function, energy metabolism and stress responses in both stromal and hematopoietic cells, providing new insights into niche regulation and cellular resilience.

Keywords: bone marrow; hematopoietic cells; stromal cells; complosome; mitochondrial metabolism

P-10: The effect of leaf extracts from *Pistacia vera* and *Rhus glabra* in combination with ionizing radiation on normal and cancerous breast cells

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Radiotherapy is one of the most commonly used treatments for breast cancer, utilizing ionizing radiation to induce DNA damage in cancer cells, leading to inhibition of proliferation and induction of cell death. The primary goal of this therapy is to maximize tumor cell destruction while minimizing damage to surrounding healthy tissues. However, the high adaptive capacity of cancer cells, their ability to develop resistance, and initiate recurrence highlight the need for more precise and personalized therapeutic strategies.

To enhance the effectiveness of radiotherapy and increase proapoptotic effects in cancer cells while protecting normal tissues, plant-derived compounds have gained increasing attention. Extracts from *Pistacia vera* and *Rhus glabra* are rich sources of bioactive compounds capable of modulating cellular responses to radiation-induced stress, including apoptosis, oxidative stress, and DNA repair mechanisms.

The aim of this study was to evaluate the effects of these extracts on breast cancer cells (MCF-7) and normal breast epithelial cells (MCF-10A). Western blot analysis was used to assess changes in protein levels associated with DNA damage and antioxidant response. Cytotoxicity was determined using the MTT assay, and IC₅₀ values were established for both cell lines.

The study focused on histone H2AX and its phosphorylated form (γ -H2AX), a marker of DNA double-strand breaks, as well as antioxidant-related proteins, including SOD1, SOD2, catalase, and GPX1/2 and GPX4. Additionally, NRF2 levels were evaluated as a key regulator of cellular antioxidant defense, with β -actin used as a reference protein.

The results indicate that the tested extracts exert a dual effect on cellular response to oxidative stress, enhancing DNA damage while simultaneously increasing the expression of proteins involved in reactive oxygen species detoxification and cellular protection.

Keywords: radiotherapy, oxidative stress, *Pistacia vera*, *Rhus glabra*

P-11: The TRPA1 channel differentially regulates calcium signalling in phenotypically distinct human pancreatic stellate cells

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The transient receptor potential ankyrin 1 (TRPA1) channel is a non-selective cation channel involved in cellular responses to chemical and physical stimuli, including the regulation of intracellular calcium levels. It is expressed in the plasma membrane of pancreatic stellate cells (PSCs). PSCs are key regulators of pancreatic tissue homeostasis, yet their persistent activation contributes to fibrosis under pathological conditions such as chronic inflammation and cancer.

This study examined TRPA1-dependent calcium signalling in human PSCs (hPSCs) representing three phenotypic states: quiescent (qhPSC), spontaneously activated (shPSC), and TGF- β -induced (ahPSC). Channel activity was pharmacologically manipulated using the agonist ASP7663 and the antagonist A-967079, while intracellular calcium dynamics were monitored using live-cell fluorescence imaging.

TRPA1 activation evoked robust calcium responses predominantly in qhPSCs, whereas both activated phenotypes displayed markedly attenuated signalling. This reduced responsiveness was associated with lower TRPA1 expression, as confirmed by qPCR and immunofluorescence analyses. Notably, antagonist application after channel activation did not fully suppress calcium signals, while preincubation with A-967079 effectively prevented their induction. Preliminary binding site modelling analyses suggest that both the agonist and antagonist may potentially interact with the same binding pocket.

Overall, the data identify TRPA1 as a potential modulator of calcium signalling in PSCs and support its further investigation in the context of pancreatic fibrosis.

Keywords: TRPA1, pancreatic stellate cells, calcium signalling

P-12: Uncovering Selective Anticancer Potential of Cyanobacterial Metabolites from *Nostoc edaphicum* CCNP1411 Against Neuroblastoma

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Neuroblastoma, also known as sympathetic neuroectodermal tumor, is one of the most common solid tumors in children and is responsible for approximately 15% of cancer-related deaths in this age group. It is characterized by rapid growth, extensive metastasis, and nonspecific symptoms, making early diagnosis and effective treatment a significant therapeutic challenge. Most patients are diagnosed at an advanced stage of the disease, and recurrence occurs in nearly half of cases. Neuroblastoma exhibits unique features, such as early age at onset and a tendency for spontaneous regression in infants, with clinical outcomes largely determined by tumor biology.

Cyanobacteria inhabiting unique environments, such as the Baltic Sea, represent a rich source of structurally diverse metabolites with potential therapeutic properties. The aim of this study was to evaluate the anticancer activity of metabolites isolated from the Baltic cyanobacterial strain *Nostoc edaphicum* CCNP1411 against neuroblastoma cells and to assess their selectivity toward normal cells.

Human neuroblastoma cell lines (SH-SY5Y and IMR-32) and normal human dermal children fibroblasts (HDFc) were used as *in vitro* models. Cell viability following treatment with the tested metabolites was assessed using the MTT assay across a range of concentrations. The analyzed compounds exhibited a dose-dependent cytotoxic effect on neuroblastoma cells, while their impact on normal fibroblasts was less pronounced, suggesting potential selectivity toward cancer cells.

These findings indicate that metabolites derived from *Nostoc edaphicum* CCNP1411 may represent promising candidates for further investigation as novel anticancer agents. Ongoing studies are focused on elucidating the molecular mechanisms underlying their cytotoxic activity.

Keywords: Neuroblastoma; Cyanobacterial metabolites; Baltic cyanobacteria; Anticancer activity; Selective cytotoxicity

P-13: Enhancing the nutritional profile of pasta dough through microencapsulated polyphenols

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Introduction:

Pasta is a widely consumed cereal product and an effective carrier of bioactive compounds including polyphenols. However, polyphenols are sensitive to processing conditions, therefore, microencapsulation is applied as a strategy to improve their stability and enable their effective incorporation into functional foods.

Objective:

The aim of this study was to develop and optimize pasta enriched with microencapsulated polyphenols, oat fiber, and sauerkraut juice, and to evaluate its physicochemical and sensory properties.

Materials and Methods:

Blackcurrant polyphenols were microencapsulated using maltodextrin via freeze-drying. Pasta formulations were designed using response surface methodology, with varying levels of microcapsules (0–18%), oat fiber (0–10%), and sauerkraut juice (0–30%). Rheological, textural, color, and nutritional parameters (TPC, TFC, TAA, vitamin C, β -glucan) as well as consumer acceptance were analyzed.

Results:

Microcapsules significantly increased total polyphenols, antioxidant activity, and vitamin C level, while also affecting color and texture. Oat fiber enhanced viscosity, hardness, and β -glucan content. Sauerkraut juice contributed mainly to vitamin C enrichment. The optimized formulation (11.59% microcapsules, 6.12% oat fiber, 11.93% juice) showed high bioactive compound content (TPC 127.81 mg GA/100 g; TAA 34.12%), acceptable texture, and good sensory acceptance (5.82). Model predictions were consistent with experimental results ($R^2 > 0.85$).

Conclusions:

The incorporation of microencapsulated polyphenols, oat fiber, and sauerkraut juice enables the development of nutritionally enhanced pasta with maintained quality and consumer acceptance.

Keywords: functional food, polyphenol microcapsules, oat fiber, sauerkraut juice, response surface methodology

P-14: TheraChip – development of a simulation-supported microfluidic lab-on-a-chip for 3D spheroid culture for personalized cancer photodynamic therapy

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Photodynamic therapy (PDT) is a selective therapeutic method that utilizes light-activated photosensitizers. After light absorption, these molecules enter an excited state, leading to the generation of reactive oxygen species. The strong oxidative properties of the generated reactive oxygen species induce targeted cancer cell death, limiting damage to surrounding healthy tissues. Despite its clinical success optimizing PDT requires researching new photosensitizers that are even more efficient and selective.

Nowadays, most of the studies on new photosensitizers are conducted using simple, two-dimensional cell cultures (monocultures), which fail to capture the complexity of the tumor environment. To better reflect the three-dimensional in vivo tumor microenvironment and its heterogeneous cellular composition (including both cancer cells and various types of healthy cells that form the tissue and vascular stroma of the tumor), a microfluidic lab-on-a-chip device was developed for spheroid culture.

This system enables the parallel formation of uniform spheroids, reducing variability between replicates and allowing for the generation of a sufficient number of technical replicates within a single microfluidic experiment. Furthermore, it will be used to analyze the accumulation of novel, previously untested photosensitizers, their cytotoxicity, and the effects of PDT on healthy cells.

This work outlines the preliminary phase of the research, focusing on computer flow simulations and the optimization of cell culture techniques for spheroid formation within the microfluidic device, and demonstrating a proof-of-concept for PDT.

Scientific work funded by the program of the Minister of Science and Higher Education entitled "Student Research Groups Create Innovations," project no. SKN/SP/658036/2026, with a project funding amount of PLN 69 700.

Keywords: lab-on-a-chip; 3D spheroid-based cell model; photodynamic therapy; cancer; microfluidics

P-15: Computational design of Dengue virus NS2B/NS3 protease inhibitors

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Dengue virus (DENV, Flaviviridae family) infects 100-400 million people annually, with half of the global population living in endemic regions. Despite the risk of fatal complications, no specific antiviral treatment exists. Available vaccines face challenges due to Antibody-Dependent Enhancement (ADE), which can exacerbate symptoms, highlighting an urgent need for novel, targeted therapeutics. This study focuses on designing inhibitors for the NS2B/NS3 protease, which is essential for the DENV replication cycle. Virtual Screening for compounds similar to known inhibitors was performed, identifying lead candidate for optimization.

Initially, the allosteric site was selected as the primary target to bypass the challenge associated with orthosteric inhibition, which is the relatively flat topography of the active site. Additionally, the specific structural requirements for the NS2B cofactor to maintain the protease in its active conformation were used to select the binding site. Rational redesign aimed at enhancing affinity unexpectedly yielded ligands with increased potency toward the catalytic active site. Based on ADME predictions and binding scores, three redesigned ligands were selected for molecular dynamics (MD) simulations. MD analysis confirmed stable binding within the active site for all ligands, marking them as strong candidates for experimental validation. Notably, one of the ligands exhibited a unique behavior, migrating from the active site to the allosteric pocket in the protein's inactive state. This suggests a dual-mode inhibitory mechanism: directly blocking the active site in the functional enzyme and preventing activation by stabilizing the allosteric site in the inactive conformation.

Keywords: Dengue virus; Molecular Dynamics; Drug design; NS2B/NS3 protease

P-16: Illuminating parasite immunology: Successful bacterial expression of a fluorescent *Toxocara canis* cystatin

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Parasitic nematodes, such as *Toxocara canis*, secrete various immunomodulatory proteins, including cystatins, to evade host immune responses. Understanding their biochemical properties is crucial for both parasitological research and the potential development of novel therapeutics. However, studying these proteins requires an efficient and reliable recombinant expression system.

The objective of this study was to design a construct and establish a bacterial expression system for *T. canis* cystatin. The protein was fused with Green Fluorescent Protein (GFP) primarily to enable future visualization of its cellular localization, and equipped with His-tags at both the N- and C-termini to facilitate subsequent affinity chromatography purification. The genetic construct was introduced into an *Escherichia coli* expression system. Following culture growth and induction, bacterial cells were harvested and lysed. To verify the presence of the recombinant protein, the lysates were analyzed by SDS-PAGE and Western blotting utilizing anti-His tag antibodies.

The analysis revealed a specific signal corresponding to the expected molecular weight of the GFP-cystatin fusion protein, confirming its successful biosynthesis in the heterologous bacterial host. Crucially, the recombinant protein was obtained in a soluble form, without the formation of inclusion bodies. As the expression of parasitic proteins in bacterial systems can often be challenging, confirming the presence of the intact, soluble fusion protein is a critical milestone. Future work will focus on developing a robust purification protocol utilizing the incorporated His-tags, followed by functional assays to evaluate the inhibitory activity of the recombinant cystatin. Ultimately, this expression strategy provides a valuable tool for further investigation of nematode-derived immunomodulators.

Keywords: *Toxocara canis*; cytokines; GFP; recombinant protein expression

P-17: Hemp meal as an alternative to fish meal in European perch (*Perca fluviatilis*) aquaculture

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The high reliance on animal-derived protein increases the cost and environmental footprint of European perch production. We evaluated hemp meal (HM) as an alternative protein source in extruded diets for perch reared in a recirculating aquaculture system (RAS). Juvenile perch (initial body weight 68.1 g) were assigned to four dietary treatments (HM0, HM10, HM20, HM30; n = 100 per treatment) and fed diets containing 0, 10, 20, or 30% HM for 10 weeks. HM inclusion did not adversely affect growth performance, muscle proximate composition, or basic blood parameters, and no severe histopathological lesions were observed. Performance indices (SGR, FCR, PER) were highest in the HM20 group. Intestinal histology in HM-fed fish showed predominantly adaptive features, and gene-expression patterns in the intestine and liver differed between higher (HM20–30) and lower/no (HM0–10) HM inclusion. Hepatic alkaline phosphatase (ALP) and glutathione peroxidase (GPX) activities decreased with increasing HM inclusion. Overall, HM can partially replace fishmeal without compromising digestive organ homeostasis, with 20% HM appearing the most promising inclusion level under the conditions tested.

*This study was a part of the project titled “Diversification of the productive function of earthen ponds based on semi-intensive rearing of *Perca fluviatilis*—PROPERCH” (project no. 00002-6521.1-OR1400004/17/20), funded by the European Union through the Operational Program “Fisheries and Sea (2014–2020)”, The Agency for Restructuring and Modernization of Agriculture (ARMA) of Poland.*

Keywords: fishmeal substitute; gene expression; hemp protein; intestinal histology; plant protein.

P-18: Sensitivity of *Pseudomonas donghuensis* P482 to sodium nitroprusside- and S-nitrosoglutathione-induced nitrosative stress is influenced by the growth medium

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Nitric oxide (NO) plays an important role in mammals, plants, and microorganisms. Its functions are diverse, ranging from regulation of blood pressure, through mediation of plant responses to various biotic and abiotic stresses, to modulation of bacterial biofilm formation. NO and its derivatives, collectively known as reactive nitrogen species (RNS), can also induce nitrosative stress, damaging cellular components and potentially leading to cell death.

Although the role of NO has been well established in the symbiosis between rhizobia and leguminous plants, much less is known about its function in other plant-associated bacteria. Our previous studies demonstrated that exudates from tomato and maize roots alter the transcriptome of *Pseudomonas donghuensis* P482, increasing the expression of genes involved in the nitrosative stress response. Additionally, genes related to amino acid catabolism were upregulated. These observations suggest that nitrosative stress may play a role in plant-microbe communication and that the utilization of alternative carbon sources may represent a form of metabolic adaptation.

In this study, we determined the resistance of *P. donghuensis* P482 to nitrosative stress induced by the nitric oxide donors, sodium nitroprusside (SNP), and S-nitrosoglutathione (GSNO), measured as the minimum inhibitory concentration (MIC). We also showed that medium composition affects the sensitivity of P482 to nitrosative stress, as reflected by differences in MIC values. The obtained results provide a basis for further investigation of the mechanisms underlying nitrosative stress resistance in *P. donghuensis* P482.

Keywords: nitrosative stress; *Pseudomonas donghuensis* P482; sodium nitroprusside; S-nitrosoglutathione

P-19: Methicillin Resistance and SCCmec Diversity in *Staphylococcus pseudintermedius* Isolated from Dogs

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Staphylococcus pseudintermedius is a common part of the canine skin microbiota and an important opportunistic pathogen associated with canine pyoderma. The emergence of methicillin-resistant *S. pseudintermedius* (MRSP), mediated by the *mecA* gene carried on the staphylococcal cassette chromosome *mec* (SCC*mec*), represents a growing concern in veterinary medicine. This study aimed to determine the prevalence of *mecA* carriage and characterize SCC*mec* types among *S. pseudintermedius* isolates obtained from dogs.

A total of 55 *S. pseudintermedius* isolates collected from 51 dogs in Poland were analyzed. Samples originated from both clinically healthy dogs (n = 27) and dogs diagnosed with pyoderma (n = 24). Species identification was confirmed by PCR detection of the *nuc* gene. Methicillin resistance was assessed by PCR amplification of the *mecA* gene, and SCC*mec* typing was performed using PCR assays.

Among all isolates, 21 (38.2%) carried the *mecA* gene and were classified as MRSP. *mecA*-positive strains were detected in isolates obtained from both healthy dogs (9/27; 33.3%) and dogs with pyoderma (12/28; 42.9%). SCC*mec* typing of MRSP isolates revealed three cassette types, with SCC*mec* II–III being the most prevalent (n = 10), followed by SCC*mec* V (n = 9) and SCC*mec* IV (n = 2).

The observed prevalence of MRSP falls within the wide range reported globally. The predominance of SCC*mec* II–III and SCC*mec* V is consistent with previously reported epidemiological trends in canine *S. pseudintermedius*. These findings highlight the circulation of multiple SCC*mec* elements within canine populations and emphasize the importance of continued surveillance of MRSP in veterinary medicine.

Keywords: *Staphylococcus pseudintermedius*; *mecA*; SCC*mec*; MRSP; methicillin resistance

P-20: Accelerated prototyping of recombinant proteins using a transient expression system in *Nicotiana tabacum* BY-2 cell packs

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Introduction

The recombinant protein market is rapidly expanding. Mammalian and bacterial systems are the current industrial benchmarks. However, plant-based transient expression is becoming recognized as a fast, scalable, and animal-free alternative.

Aim of the study

We evaluated the utility of *Nicotiana tabacum* BY-2 plant cell packs (PCPs) as a high-efficiency matrix for transient recombinant protein synthesis.

Material and methods

Gene encoding avGFP was cloned into a pEff-based vector (AscI/XmaI), electroporated into *Agrobacterium tumefaciens* strain GV3101, and introduced into BY-2 cultures *via* vacuum infiltration. Expression was analyzed after 3–5 days by microscopy and Western blot.

Results

The system successfully produced avGFP. Proteomic analysis suggested protein yields much higher than those in stably transformed lines. Substantial accumulation occurred within only 72 hours. This provides a major time advantage over conventional methods, which require weeks of selection.

Conclusion

This study shows that plant cell packs accelerate production and maintain high yields. Therefore, this platform is well-suited for rapid prototyping of biopharmaceuticals. Future research will concentrate on increasing throughput and automating the process.

Keywords: Protein Recombinant expression plant transient

P-21: How to Deliver a Peptide Across the Skin? A Case Study of BPC-157

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BPC-157 is a pleiotropic pentadecapeptide (GEPPPGKPADDAGLV), originally isolated from human gastric juice as a fragment of the Body Protection Compound (BPC). It has attracted considerable attention due to its broad cytoprotective and regenerative effects, as well as its stability under acidic conditions[1]. However, peptides are generally poor drug candidates, and their oral or transdermal administration is typically inefficient. Consequently, injectable formulations remain the dominant route of administration. In transdermal delivery, the main limitations include the lipophilic nature of the stratum corneum, high molecular weight, hydrophilicity, and the presence of charge[2].

The aim of this study was to develop a transdermal formulation of BPC-157 using two complementary strategies: lipidation with palmitic acid and encapsulation in transferosomes (elastic liposomal carriers that enhance penetration through the stratum corneum). Lipidation increases peptide lipophilicity, improves its affinity for biological membranes, and prolongs its half-life (e.g., via transient binding to albumin). Encapsulation, in turn, protects the peptide from degradation and facilitates its transport across the skin barrier[3,4].

The study involved the synthesis of BPC-157 via solid-phase peptide synthesis, followed by lipid modification and encapsulation in lipid-based nanocarriers. The structure of the obtained peptides and the efficiency of lipidation were confirmed using LC-MS. The physicochemical properties of transferosomes were characterized by dynamic light scattering, including hydrodynamic diameter and polydispersity index, while zeta potential measurements were performed to assess their electrokinetic stability.

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Keywords: BPC-157; skin permeability; lipidation; encapsulation; transferosomes

P-22: Sensor Arrays for Rapid Genetic Point-of-Care Diagnostics: Optimization of DNA Probes for Bacterial Identification

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As **antimicrobial resistance** becomes one of the greatest challenges of 21st-century medicine, the shift from centralized laboratory testing to rapid, on-site diagnostics is no longer a luxury, but a necessity.

This work is part of a project developing sensor arrays for rapid genetic analysis in automated **Point-of-Care (PoC)** devices. In this part, we focus on the electrochemical detection of *ddlEfm* gene (108 bp) for the identification of *Enterococcus faecium*, a bacterium known for its resistance to many commonly used antibiotics.

Three DNA probes were designed for target gene to optimize hybridization efficiency. The detection mechanism was based on DNA probes and complementary methylene blue-labelled DNA targets. The current responses was generated only after the hybridization reaction, enabling selective gene detection. Sensors were evaluated using Square Wave Voltammetry (SWV). We analyzed synthetic targets at 50nM and 200nM concentrations, alongside non-complementary sequence (200nM). The best-performing probes were validated using purified real-world samples obtained via RPA.

The optimal probe was selected based on the highest current response after 30 min hybridization time, showing minimal cross-reactivity. Thermodynamic analysis revealed that probes with higher accessibility – characterized by higher ΔG_{max} value (related to lower secondary structure stability) - yielded superior signals. Interestingly, the ease of probe unfolding proved more critical for signal magnitude than the proximity of the redox label to the electrode. Furthermore, varying primer ratios during amplification directly influenced the final current response.

Optimizing probe thermodynamics and amplification conditions **significantly improves biosensor reliability**. This enables the integration of sensitive sensor arrays into automated diagnostic platforms, paving the way for faster and **more precise clinical decision-making**.

Keywords: Electrochemical biosensor; DNA probe design; Point-of-Care diagnostics; Thermodynamics; Bacterial identification

P-23: Cellular response to DNA damage induced by 405 nm laser irradiation

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Exposure to UV radiation induces DNA damage, primarily cyclobutene pyrimidine dimers and 6-4 photoproducts. While visible light is known to induce single-strand breaks (SSBs)[1], radiation at 405 nm – positioned at the boundary between the visible and UV spectra – generates damage characteristic of both ranges[2].

The goal of this work was to determine the dose of 405 nm pulse mode laser radiation required to induce SSBs. DNA lesions were generated in HeLa cells expressing mRFP-XRCC1 protein by micro-irradiating a selected region of the cell nucleus. Since XRCC1 plays a crucial role in the BER (Base Excision Repair) and SSB (Single-Strand Break Repair) pathways[3], its accumulation at the site of irradiation site served as a marker of induction of DNA damage.

The results demonstrate that the probability of SSB induction increases linearly with the delivered energy, reaching a value of 1 for energies exceeding 1.2 μJ . Furthermore, preliminary data indicate that the 405 nm beam is also capable of inducing double-strand breaks (DSBs). These findings contribute to a better understanding of the mechanisms underlying light-induced DNA damage and repair. In addition, they may find application in studies requiring the controlled induction of specific DNA lesions within living cells.

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Keywords: DNA single-strand breaks (SSB); Light-induced DNA damage; XRCC1

P-24: Electrochemical tongue – a sensor array for the detection of date-rape drugs (DRDs) and other pharmaceuticals

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The aim of a project (NCN 2023/50/E/ST4/00639) is to develop an electrochemical tongue – sensor array for the detection of **date-rape drugs** and other pharmaceuticals. The problem of drug-facilitated crimes is a deeply misunderstood issue, from under-reporting to analytical difficulties due to complex samples and their matrices. Development of a portable sensor will aid the potential victims in the prevention and the police during the first response. Electrochemical methods are perfect for this due to their analytical capabilities, as well as a relatively simple setup and the possibility of miniaturisation. The development of a sensor opens the door not only for the forensic sector, but also for other applications like real-time monitoring in bioreactors, in vivo cell culture control and many similar, biotech and beyond. The project distinguishes itself by the integration of machine learning to extract the most out of the simplest systems.

Chosen DRDs are γ -hydroxybutyric acid, its precursors, and benzodiazepines – alprazolam (Xanax) and flunitrazepam (Rohypnol). Codeine is investigated too, as it is available as an OTC medicine in Poland, and is prone to at-home abuse. The first step is the selection of working electrode materials. The testing involves low-cost paper graphite electrodes drawn with pencils, 3D printed electrodes from PLA/carbon black and PLA/graphene filaments, and PLA/carbon composites using materials such as nanotubes and recycled coffee beans, grinded together and remelted into silicon molds.

The preliminary results of cyclic voltammetry (CV) and differential pulsed voltammetry (DPV) were measured in two different 3-electrode setups – a 20 μ l droplet on a working electrode, or with paper as an interface with screen-printed reference and counter electrode. Codeine distinction from paracetamol, its interferent, is promising using just CV. The distinction between benzodiazepines can also be seen when DPV is applied, as it is more sensitive.

Keywords: electrochemical tongue; sensor arrays; carbon nanomaterials; 3d printing; low-cost

P-25: When intracellular calcium is not enough: extracellular influx drives T Cell activation

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Calcium signaling is a key regulator of T-cell activation, enabling the sustained increase in intracellular Ca^{2+} concentration necessary for calcineurin activation, nuclear translocation of Nuclear Factor of Activated T-cells (NFAT), and the induction of cytokine expression. The current model posits a dominant role for the store-operated calcium entry (SOCE), initiated by depletion of Ca^{2+} stores in the endoplasmic reticulum and activation of Orai1 channels by STIM1. However, it remains unclear whether disturbances in calcium homeostasis within organelles can functionally regulate NFAT activation independently of extracellular influx.

The aim of this study was to determine whether modulation of intracellular Ca^{2+} can induce NFAT activation in T lymphocytes. The study utilized Jurkat-LuciaTMNFAT reporter cells treated with verteporfin, brefeldin A, and ochratoxin α - compounds that disrupt vesicular transport, ER and mitochondrial function, and SERCA pump activity. Analyses were conducted in the presence and absence of extracellular Ca^{2+} and following stimulation with PMA, PHA, and ionomycin, a calcium ionophore. NFAT activity and cell viability were assessed with QUANTI-LucTM and MTT assays.

Despite the varied effects of the tested compounds on cell survival and activation levels under stimulation conditions, the key observed effect was a marked reduction in NFAT activation in a Ca^{2+} free environment. Even strong stimulation failed to restore high activity of the transcription factor. The results indicate that the release of Ca^{2+} from intracellular stores does not compensate for the lack of extracellular influx, highlighting the fundamental role of SOCE in regulating T-cell responses.

The study provides new evidence suggesting that organelle dysfunction modulates, but does not replace, key mechanisms controlling NFAT signaling.

Keywords: Calcium signaling; NFAT; T-cell activation; Store-operated calcium entry; Organelle stress

P-26: Alginate Hydrogel-Based Delivery of Antimicrobial Peptides: Controlled Release and Preserved Antibacterial Activity

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The rapid emergence of antibiotic-resistant bacteria represents a critical global health challenge, driving the search for innovative antimicrobial strategies. Antimicrobial peptides (AMPs) have gained increasing attention due to their broad-spectrum activity and low propensity to induce resistance. However, their clinical translation requires effective delivery systems ensuring stability, controlled release, and sustained antimicrobial efficacy.

Alginate, a natural polysaccharide derived from brown algae, is a biodegradable, biocompatible, non-immunogenic, and cost-effective biomaterial. Its ability to form hydrogels under mild conditions makes it particularly suitable for encapsulating sensitive therapeutic molecules such as AMPs without compromising their activity. Additionally, the tunable porous structure of alginate matrices enables control over release kinetics while limiting premature degradation.

In this study, alginate hydrogels were developed as carriers for antibacterial peptides with lysine- and leucine-rich sequences, and their release profiles were investigated. Peptide release was quantitatively monitored via intrinsic tryptophan fluorescence using a microplate reader, enabling real-time tracking of diffusion from the hydrogel matrix. By modulating system parameters, optimal release conditions were identified. The resulting profiles showed that hydrogels maintain AMP concentrations above the minimal inhibitory concentration (MIC) for up to two days. Furthermore, bacterial culture assays evaluating AMP-loaded hydrogels will be presented to assess their ability to inhibit bacterial growth.

Overall, these results demonstrate that alginate hydrogels represent a versatile and promising platform for controlled AMP delivery, with potential applications in combating antibiotic-resistant infections.

Keywords: Antimicrobial peptides (AMPs); Alginate hydrogels; Controlled release; Drug delivery systems; Antibiotic resistance

P-27: RyhB1 and RyhB2 sRNAs shape Iron-Dependent Regulatory Networks in *Yersinia enterocolitica*

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Yersinia enterocolitica exhibits a dual lifestyle, thriving both as a non-pathogenic saprophyte and as a gastrointestinal enteropathogen. This dual lifestyle requires the bacterium to rapidly adapt to diverse environmental conditions encountered both in external environments and within the host. Small regulatory RNAs (sRNAs) play a crucial role in bacterial regulatory networks and stress responses, influencing survival and adaptation. Among them, the sRNA RyhB is regulated by the Fur protein and plays a central role in iron metabolism. In the genus *Yersinia*, two paralogs of this sRNA are present: RyhB1 and RyhB2.

This study aimed to perform the molecular characterization of the sRNAs RyhB1 and RyhB2 and to assess their regulatory effect on the expression of selected genes in *Y. enterocolitica*.

Experiments were performed using *Y. enterocolitica* strains with different levels of RyhB1 or RyhB2 activity. Potential transcription start sites of the genes encoding these sRNAs were identified *in silico* and experimentally confirmed by Primer Extension analysis. Bioinformatic analyses using IntaRNA were performed to predict potential base-pairing interactions between RyhB1/RyhB2 and the mRNAs of selected genes. Gene expression levels were determined by RT-qPCR.

Experimental data confirmed the predicted transcription start sites of RyhB1 and RyhB2 and showed that their expression is dependent on iron availability. Gene expression analyses indicated that both sRNAs regulate multiple genes involved in stress responses, motility, iron acquisition, biofilm formation, and virulence in *Y. enterocolitica*.

These findings highlight the role of RyhB1 and RyhB2 in iron-dependent regulatory networks in *Y. enterocolitica* and contribute to a better understanding of the mechanisms underlying bacterial adaptation, survival, and pathogenicity.

This work was supported by the National Science Center, Poland (UMO-2019/33/N/NZ1/00484).

Keywords: *Yersinia enterocolitica*; sRNA; RyhB1; RyhB2; gene expression regulation

P-28: Effects of use of Foliar Silica on growth of *Vitis vinifera* L. Grapevine

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Silicon (Si) is recognised as a beneficial element that enhances plant tolerance to various environmental stresses, yet its physiological and molecular roles in perennial fruit crops such as grapevine (*Vitis vinifera* L.) are not fully understood. In the context of organic farming, where the use of natural amendments is prioritized, silica applications, particularly in the form of nanoparticles, offer a promising and sustainable strategy for improving plant performance and fruit quality. This study investigates the effects of foliar-applied silica nanoparticles (SiNPs) on the growth and photosynthetic efficiency of grapevines cultivated under organic field conditions.

Field experiments are being conducted at the certified organic Wieliczka Vineyard in southern Poland, where several *V. vinifera* cultivars, including 'Cabernet,' are grown. Grapevines were treated with silica nanoparticles through foliar spraying at key stages of vegetative development, while untreated served as controls. All plants were grown under uniform agronomic practices, ensuring that observed responses could be attributed to the SiNP treatment. Throughout the growing season, non-destructive physiological measurements were performed to evaluate the effects of SiNP application on grapevine performance.

Physiological assessments included net photosynthesis, stomatal conductance, transpiration, and chlorophyll fluorescence to evaluate photosystem II performance. Leaf area and pigment content were measured, while soil properties and weather data were analysed to characterise baseline conditions.

Preliminary results show that grapevines treated with SiNPs exhibit measurable differences in several photosynthesis-related parameters compared with untreated controls. These observations suggest that foliar SiNP application may alter grapevine leaf physiology under organic field conditions. Although the mechanisms remain unclear, the findings provide field-based evidence that SiNPs influence performance.

Keywords: Nanoparticles; silica nanofertiliser; plant stress resistance; organic farming

P-29: Characterization of a Newly Isolated Bacteriophage 4.1 Targeting *Salmonella enterica*

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Increasing bacterial antibiotic resistance is one of the major challenges in modern medicine, reducing the effectiveness of current methods used to treat and control infections. This issue is especially important in *Salmonella*, one of the most common zoonotic pathogens causing gastrointestinal infections in both humans and animals. The common occurrence of *Salmonella* serotypes contributes to the persistence of pathogen reservoirs and increases the selective pressure associated with antibiotic use. In this context, alternative and precise antibacterial strategies, such as phage therapy, are gaining particular importance. Phage therapy uses bacteriophages as natural, highly specific agents against bacterial pathogens.

The aim of this study was to characterize newly isolated bacteriophage 4.1 capable of infecting and lysing *Salmonella* cells. Its physicochemical properties were evaluated, including stability over a wide range of pH values and temperatures. In addition, host range, efficiency of plating (EOP), adsorption kinetics, one-step growth, and lytic activity against selected *Salmonella enterica* strains were determined.

Bacteriophage 4.1 showed high stability over a pH range of 4–12 and thermal stability up to 60°C. Its host range included selected strains representing 100 *Salmonella enterica* serotypes relevant to salmonellosis. EOP analysis demonstrated lytic efficiency against 46% of tested strains. Furthermore, the analysis of adsorption kinetics and the one-step growth curve showed that the phage exhibited rapid and efficient adsorption to the tested strains, while at 37°C the burst size was observed after 7.5 minutes and extended until 20 minutes.

These results suggest that bacteriophage 4.1 has therapeutic potential as a component of phage-based preparations intended for the prevention and treatment of *Salmonella* infections. Further studies, including genomic analysis and *in vivo* testing, are necessary to fully evaluate its applicability in phage therapy.

Keywords: bacteriophage, antibiotic resistance, *Salmonella*, phage therapy

P-30: Integrated mRNA and miRNA profiling of the hermaphroditic Hgy3 cucumber line

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Cucumber (*Cucumis sativus*) serves as a model species for investigating the processes related to sex determination in plants. It exhibits a complex mechanism of sexual differentiation, which is a highly coordinated and genetically regulated process. While the development of flower organs is known to be regulated at both mRNA and microRNA (miRNA) levels, the precise interactions governing these processes remain incompletely understood. The hermaphroditic Hgy3 cucumber line produces both pistils and stamens within a single flower bud. The flower buds of this line are characterized by shorter and more variable ovary length compared to buds observed exclusively in pistillate flowers.

This study aimed to elucidate regulatory pathways involved in flower development by analyzing differentially expressed genes (DEGs) identified via RNA sequencing (RNA-seq) of flower buds, leaves and shoot apices from the hermaphrodite cucumber line Hgy3. In parallel, small RNA (sRNA) sequencing was employed to identify differentially expressed miRNAs and their corresponding target genes. Gene ontology (GO) enrichment analysis was performed to study the molecular functions and biological processes associated with these genes. Additionally, the obtained expression profiles were compared with those of the reference B10 line, a male cucumber line, to identify key differences associated with sex-specific developmental pathways and to better understand the genetic basis of floral sex differentiation.

These findings provided new insights into the complex genetic and epigenetic mechanisms underlying floral development.

This research was funded by a project from the National Science Center UMO-2020/37/B/NZ9/00586

Keywords: cucumber; transcriptome; miRNA; sex differentiation

P-31: Verification of the effectiveness of magnetic nanoparticle-mediated photothermal therapy in a lab-on-a-chip system

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Nowadays, cancer is the second leading cause of death worldwide, following cardiovascular diseases. Currently applied therapeutic strategies are often insufficiently effective and frequently associated with significant side effects. Therefore, there is a pressing need for novel cancer treatment methods that are both effective and minimally invasive, while reducing harmful side effects.

One promising approach is photothermal therapy, which induces cancer cell death through a localized increase in temperature. This temperature rise results from the absorption of laser radiation by nanoparticles accumulated within tumour tissue. Consequently, for this therapy to be effective, its key parameters—such as irradiation time, wavelength, laser power, and type of nanoparticles—must be carefully optimized.

In this study, a dedicated microsystem was designed and fabricated to facilitate both controlled irradiation of cells and rapid assessment of metabolic activity changes using a multiwell plate reader. Moreover, the incorporation of multiple independent microchambers enables the simultaneous analysis of different experimental conditions, including control samples without irradiation, irradiated samples, as well as samples containing nanoparticles with and without photoactivation.

The developed microsystem was used to determine the effectiveness of photothermal therapy employing magnetic nanoparticles ($\text{Fe}_3\text{O}_4@Au$ DOX-BSA) in HaCaT (human keratinocytes) and A375 (melanoma) cell models. Therapeutic efficacy was evaluated using metabolic activity measurements (Alamar Blue assay). The system provides a platform for assessing the efficacy of nanoparticles as photoactive agents and for optimizing photothermal therapy protocols.

Keywords: lab-on-a-chip, photothermal therapy, magnetic nanoparticles, energy conversion, cancer cells

P-32: Redefining the biosynthetic potential of *Aralia spinosa* L. – establishment and phytochemical profiling of hairy root cultures

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Aralia spinosa L. is a medicinal species of the Araliaceae family, traditionally used against rheumatism and in skin-related remedies due to its anti-inflammatory properties. It is regarded as a source of specialized metabolites, particularly triterpenoid saponins. However, *A. spinosa* has not previously been explored as a hairy root culture system. The aim of this study was to establish hairy roots of *A. spinosa*, confirm transformation, select representative lines and evaluate its biosynthetic potential as a source of bioactive metabolites in comparison with anatomical roots of seedlings cultivated *in vitro*. Hairy roots were induced in *in vitro*-growing plantlets using *Rhizobium rhizogenes* strain A4. The resulting lines underwent selection, molecular verification and qualitative phytochemical characterization. PCR analysis confirmed stable transformation of the selected lines through the presence of genes: *rolA*, *rolB*, *aux1*, *mas1*, and *ags1*, while the absence of *virG* and *virD1* indicated the lack of residual bacterial contamination. The roots displayed a typical hairy root phenotype, with line-specific differences in growth characteristics and branching pattern.

Qualitative UHPLC-DAD-ESI-MS3 analysis of methanolic extracts revealed a metabolite profile characteristic of the genus *Aralia*, dominated by triterpenoid saponins and phenolic acids. Identified constituents included araloside A, calenduloside E, chlorogenic acid and isochlorogenic acid A. The transformed roots retained the major secondary metabolites detected in anatomical roots while offering advantages typical of hairy root cultures, including stable and rapid hormone-independent growth.

To the best of our knowledge, this is the first report on the establishment of hairy root cultures in *A. spinosa*. The results demonstrate the susceptibility to agroinfection and indicate that the obtained cultures represent a promising *in vitro* platform for the controlled production of valuable plant secondary metabolites.

Keywords: *Aralia spinosa*; hairy root culture; genetic transformation; specialized metabolites; phytochemical profiling

P-33: Screening of the *Pseudomonas donghuensis* P482 miniTn5 mutant library for its ability to form biofilms on abiotic surfaces and plant tissues.

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The use of chemical agents and fertilizers in agriculture contributed to high levels of environmental pollution. Therefore new, alternative methods of plant protection and growth promotion are intensively investigated into. Application of Plant Growth-Promoting Rhizobacteria (PGPR), beneficial bacteria from rhizosphere, is a developing method allowing to improve plant's growth and crop yield or protect plants against pathogens. *Pseudomonas donghuensis* P482 is a potential biocontrol agent isolated from tomato rhizosphere in Gdynia, Poland. It inhibits the growth of bacterial and fungal pathogens such as *Pectobacterium spp.*, *Dickeya spp.*, or *Fusarium sp.* P482 efficiently colonizes tomato, maize and potato rhizosphere, what is important in competition with pathogenic microorganisms. The colonization of plant tissues and biofilm formation are multistep processes highly regulated by internal and external factors which can differ depending on the type of surface. Earlier research on P482 showed that mutations in biofilm formation related genes impact biofilm formation ability on abiotic but not on biotic surfaces. The exact molecular mechanisms behind root colonization in this strain are little understood. In this study a library of *P. donghuensis* P482 miniTn5 transposon mutants was screened for strains affected in biofilm formation and/or root colonization ability. First, bacterial colony morphology on Congo-Red medium and biofilm formation on polystyrene 96-well plates under different nutritional conditions were analyzed. Selected mutants were GFP-tagged and used in assay for biofilm formation ability on glass in the previously tested conditions. Also, short- and long-term colonization of tomato roots was assessed to verify the correlation between biofilm formation on abiotic and biotic surfaces. The results of this study may expand our understanding of genetic determinants of biofilm formation and root colonization in the beneficial *P. donghuensis* P482 strain.

Keywords: *Pseudomonas donghuensis* P482, biofilm, rhizosphere colonization

P-34: The role of interleukin 6 in the replication kinetics of selected poxviruses in mouse dendritic cells *in vitro*

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Interleukin 6 (IL-6) is a key pro-inflammatory cytokine involved in the T-cell-mediated immune responses, influencing the functions of dendritic cells (DCs) at the early stages of infection. The aim of this study was to determine the role of IL-6 in the replication kinetics of ectromelia virus (ECTV) and vaccinia virus (VACV) in murine DCs. A multilevel research model was developed and implemented, based on the standardization of temporal parameters and optimization of quantitative fluorescence imaging protocols. Extraction of numerical and scalar data was introduced, enabling objective morphometric analysis of viral factories, actin tails, and adhesive structures (podosomes and focal adhesions) using image analysis tools. It was demonstrated that VACV replicates faster and induces apoptosis more strongly in both wild-type (WT) and IL-6-deficient (IL-6^{-/-}) DCs, than ECTV, reflecting different adaptive strategies of these viruses. Regardless of IL-6, both viruses caused a rapid loss of podosomes and the formation of elongated focal adhesions, resulting in cellular immobilization. A key finding was that the absence of IL-6 significantly reduced the number of actin tails, suggesting a role for this cytokine as a mediator of cytoskeletal dynamics. The obtained results indicate that the absence of IL-6 does not affect the kinetics of the poxvirus replication cycle in DCs, but may support their spread *in vitro* through modulation of actin polymerization.

Keywords: poxviruses, dendritic cells, interleukin 6, actin cytoskeleton, podosomes, focal adhesions

P-35: Biochar Functionalized with Reject Water-Derived Humic Substances as a Sustainable Biostimulant and Protective Agent for Rapeseed against *Botrytis cinerea*

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Ensuring stable crop production is a major global challenge, given that nearly 80% of plant diseases are caused by fungi. Phytopathogenic fungi employ various strategies to colonise plants, including secreting hydrolytic enzymes that degrade plant cell walls, reducing growth and crop quality. Current methods of controlling phytopathogens rely heavily on chemical fungicides, whose prolonged use promotes the development of pathogen resistance and harms the environment. Concerns over fungicide impacts highlight the need for sustainable plant protection strategies. Biochar (BC) derived from waste biomass pyrolysis has gained attention for improving soil properties, increasing microbial activity, and enhancing plant-microbe interactions. It benefits plants by stimulating root exudation and promoting microbial colonization, thereby enhancing resistance to biotic stresses. However, its ability to directly inhibit phytopathogens remains limited. Humic substances (HSs) can suppress fungal growth and cell wall biosynthesis, but their concentrations in soil are declining globally due to organic matter depletion. Furthermore, the reliance on fossil-derived HSs is ecologically unsustainable. This study evaluates, for the first time, the potential of BC from seed husk biomass functionalized with HSs (BC-HSs), derived from a renewable source - reject water obtained from the co-digestion of sewage sludge and organic waste, as a potential biostimulant and protective agent for rapeseed against the widespread phytopathogen *Botrytis cinerea*. The results demonstrate that both BC and BC-HS enhanced rapeseed growth by 32 and 10%, respectively, compared to the untreated soil. Notably, while BC alone inhibited *B. cinerea* growth by only 7% relative to the control, the addition of HSs significantly increased its antifungal activity by 28%. These findings highlight the synergistic effect of reject water-derived HSs enrichment in enhancing both rapeseed growth and pathogen suppression.

Keywords: biochar, biocontrol potential, biostimulant, humic substances, phytopathogenic fungi

P-36: Integrated approach to biotechnological regeneration of Na-P1 zeolitic sorbent contaminated with diesel oil

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Management of mineral sorbents spent on oil spill cleanup mainly involves landfilling and incineration, which are both economically costly and environmentally unfriendly. Therefore, there is a need to develop more efficient and sustainable methods for the treatment of spent sorbents. The aim of this study was to investigate the possibility of biotechnological washing of diesel-contaminated Na-P1 zeolitic sorbent and to assess the biodegradability of hydrocarbons present in the resulting effluents.

The Na-P1 zeolitic sorbent was contaminated with diesel oil at a 1:1 mass ratio. Washing experiments were conducted in a dynamic system using various washing solutions: distilled water, 1% ethanol, 1% sodium dodecyl sulfate (SDS), 1% Triton X-100, *Bacillus sp.* ANT_WA51 broth (biosurfactant-producing) and *Bacillus sp.* W78 broth (non-producing). The surface-active properties of the solutions were evaluated using the oil spreading test, surface tension measurements, and emulsification index (E2 and E24) determination. Synthetic effluents were prepared using tap water and varying amounts of diesel oil. Biodegradation of hydrocarbons in the effluents was carried out using the BioremOil consortium in the OxiTop respirometric system.

The biosurfactant-containing broth of *Bacillus sp.* ANT_WA51 exhibited properties comparable to synthetic surfactants, reducing surface tension to 27 mN/m and stabilizing emulsions (E24 \approx 60%). Similar washing efficiencies (50–54%) were observed for all tested solutions, including distilled water. Biodegradation of hydrocarbons in the effluents reached approximately 38% after 8 days of incubation.

The results suggest that strong sorption of hydrocarbons on Na-P1 zeolite may limit the effectiveness of biosurfactant-assisted washing. Nonetheless, hydrocarbons present in the resulting effluents can be reduced through biological treatment.

Keywords: Na-P1 zeolite, diesel oil, biosurfactants, biodegradation, sorbent regeneration

P-37: Polyphenol-coated hybrid magnetic nanozymes for combined photothermal and chemodynamic anticancer therapy

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The integration of nanoparticles with complementary functionalities enables the development of advanced platforms for biomedical applications. Magnetic nanoparticles (MNPs) are particularly attractive as core materials due to their responsiveness to external magnetic fields, enabling controlled manipulation.

In this work, MNPs were functionalized with catalytically active noble-metal nanoislands and subsequently coated with polyphenol-based layers formed via nanozyme-driven polymerization. The catalytic activity of the surface nanoislands enables controlled formation of polymeric coatings, yielding stable and functional nanostructures.

Fe₃O₄ nanoparticles synthesized using coprecipitation were used as magnetic cores for Pt nanoislands. The obtained hybrid nanostructures were coated with polydopamine, poly(L-DOPA), poly(pyrogallol) and poly(gallic acid). The resulting structures exhibit broad optical absorption, particularly in the near-infrared region, while preserving their catalytic activity. These properties enable their application in combined therapeutic strategies: peroxidase-like activity is associated with reactive oxygen species generation, while strong NIR absorption allows efficient photothermal conversion.

The photothermal performance of the nanostructures was evaluated under 808 nm laser irradiation, confirming efficient light-to-heat conversion and good thermal stability. Their catalytic activity was verified using TMB-based assays, demonstrating peroxidase-like behavior. Additionally, interactions with glutathione, a key intracellular antioxidant contributing to chemodynamic resistance in cancer cells, were investigated to assess the ability of the nanostructures to influence the redox balance in a tumor-like environment.

Cytotoxicity studies indicated low intrinsic toxicity of the nanostructures, while photothermal treatment resulted in a significant reduction of cancer cell viability, highlighting their potential for combined anticancer therapy

Keywords: magnetic nanoparticles, nanozymes, polyphenols, photothermal therapy

P-38: Dissecting Hair Follicle Stem Cells Niche Dynamics at Single-Cell Resolution through BMP signaling Modulation

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Hair follicle stem cells (HFSCs) orchestrate cyclical regeneration and maintain the hair follicle homeostasis. While intrinsic signaling networks governing HFSCs' quiescence and activation have been described, how these programs integrate with and remodel the surrounding niche remains poorly understood. Our previous work identified an intrinsic oscillator within HFSCs, comprising bone morphogenetic protein (BMP) and wingless-related integrated site (WNT) signaling, as a central regulator of HFSCs' quiescence and activation; however, its influence on the niche environment remains incompletely elucidated.

In this study, we aim to investigate how BMP modulation in HFSCs affects the surrounding niche environment to maintain their stemness and regeneration potential.

To achieve this, we established inducible and reversible BMPR1A gain-of-function (GoF) and conditional BMPR1A knockout *in-vivo* model systems. In the BMPR1A-GoF mouse model, doxycycline induction at postnatal day 18 (P18) disrupted the natural progression of hair cycling, efficiently blocking entry into the anagen phase. Upon doxycycline withdrawal, hair follicles exit quiescence and begin re-regeneration of hair, as evidenced by histological analysis and immunostaining. In the BMPR1A-LoF (Loss-of-function)/conditional knockout mouse model, RU induction at postnatal day 43 (P43) showed premature activation of hair follicles, bypassing quiescence and accelerating proliferation of stem cells, which was confirmed by immunostaining for activation markers. Furthermore, to dissect the molecular and cellular interplay between HFSCs and their surrounding niche at single-cell resolution, we performed single-cell RNA sequencing experiments with hair follicles' prolonged quiescence, re-regeneration, and early activation, enabling us to characterize cellular and molecular shifts in both the model system following BMP modulation.

Keywords: Hair follicle stem cells (HFSCs), bone morphogenetic protein (BMP), single cell RNA sequencing, hair regeneration

P-39: Optimization of expression and purification of recombinant West Nile Virus NS2B–NS3 protease

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West Nile Virus (WNV), a member of the *Orthoflavivirus* genus, is a mosquito-borne pathogen responsible for outbreaks of West Nile fever. Over recent decades, its geographic distribution has widened significantly, driven by continuous climatic changes. Although the majority of infections are asymptomatic or present with only mild symptoms, in some cases, the disease can escalate into severe neuroinvasive conditions like encephalitis. The resulting long-term complications remain a serious and persistent public health concern. Along with related, clinically important flaviviruses like Zika and Dengue, WNV relies on the NS2B–NS3 protease complex to cleave the viral polyprotein into functional units. Because this proteolytic processing is essential for viral replication and the enzyme is highly conserved across the genus, the NS2B–NS3 complex has emerged as a primary target for drug development. However, the recombinant production of this enzyme in *E. coli* is frequently challenged by poor solubility and autoproteolysis.

In this study, we focused on establishing an optimized workflow to produce high-quality and active NS2B-NS3 protease suitable for future inhibitor screening. The *E. coli* BL21 strain was transformed with a WNV fusion construct (Addgene #204794) carrying a K104A mutation to prevent self-cleavage. Production was optimized by testing various parameters, including different lysis buffers, induction temperature, and duration. Following sonication and lysis, the 6×His-tagged protein was purified via immobilized metal affinity chromatography (IMAC) on nickel resin. Protease presence and purity were verified by SDS-PAGE and Western blotting. The final step involved assessing the activity of the purified protease using a fluorogenic assay.

Keywords: Flaviviruses, West Nile Virus (WNV), NS2B-NS3 Protease, Protein expression optimization

P-40: Data-driven calibration of collective event detection in spatio-temporal cell signaling

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Collective ERK signalling waves coordinate cell behaviour in space and time, but their quantitative detection remains sensitive to user-defined parameters. ARCOS identifies collective events by clustering active cells in each frame and linking clusters across time, with key parameters including the neighbourhood radius and minimum cluster size. Although practical recommendations exist, parameter choice remains partly heuristic and can affect the number, size and duration of detected events.

We propose a data-driven framework for calibrating ARCOS using both synthetic and experimental data. As an experimental testbed, we build on the ARCOS setting of ERK activity imaging in starved MCF10A epithelial cells, including wild type as well as KRAS G12V and PIK3CA H1047R mutants expressing the ERK-KTR biosensor. In this system, ARCOS revealed that oncogenic mutants display more, larger and longer collective ERK events than wild-type cells, while inhibition of matrix metalloproteinase-dependent intercellular communication with batimastat strongly reduces these events.

Using synthetic data with controlled event size, duration and noise, we quantify how ARCOS parameters influence recovery of ground-truth events and transfer these insights to experimental recordings. The goal is to derive experiment-specific calibration rules that reduce subjective tuning, improve robustness, and support reproducible comparison of collective signalling across conditions.

Keywords: ERK, signalling, calibration, spatio-temporal, simulation

P-41: Effects of Bacillus-Derived Osmoprotectants on Enzymatic Activity and Microbial Communities Composition in Saline Soils

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Soil salinity is a major stress factor limiting microbial activity, soil fertility, and agricultural productivity. Microorganisms respond to osmotic stress through the production and uptake of osmoprotectant metabolites that stabilize cellular functions under high salt concentrations. The main objectives of this study were to (i) evaluate the effect of bacterial osmoprotectants on microbial enzymatic activity and soil quality, (ii) assess the impact of supplementation frequency, and (iii) examine changes in soil microbial community structure.

Three saline soils differing in pH (acidic, neutral, and alkaline) were supplemented once or weekly with osmoprotectant metabolites derived from *Bacillus sp.*, consisting of an amino acid mixture rich in betaine (322,278 $\mu\text{M g}^{-1}$ bacterial proteins), applied alone or with bacterial cells.

Microbial activity and soil quality were assessed through soil respiration, organic matter content, and dehydrogenase activity. In neutral soil, protease and β -glucosidase activity assays were also measured. Supplementation with bacterial metabolites increased microbial activity and soil quality after single or repeated application. Soil respiration increased by up to 2,6% compared to the control, while dehydrogenase activity reached up to 72% above control depending on soil type and treatment. Bacterial cultures increased soil organic matter content from 2,04% to 2,97%. In neutral soil, β -glucosidase activity increased significantly, by up to 95%, indicating strong stimulation of carbon cycling. Contrary to protease activity which decreased compared to the control, suggesting a shift in metabolic activity.

Furthermore, full-length 16S rRNA sequencing (Oxford Nanopore) revealed shifts in microbial community structure, suggesting that metabolites alter microbial composition under saline stress.

These findings highlight the potential of bacterial osmoprotectants as a strategy to enhance microbial activity and soil quality in saline environments.

Keywords: Soil salinity, osmoprotectants, microbial activity, soil enzymes, microbial community structure

P-42: Biotransformation of *Hypericum perforatum* compounds by selected probiotic products.

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Hypericum perforatum is a well-established herbal antidepressant used in the management of mild depressive disorders. Although its clinical efficacy is supported by preclinical and clinical evidence, the mechanisms underlying its activity and the contribution of specific bioactive compounds remains incompletely understood. In particular, little is known about the role of bacteria-mediated biotransformation of *H. perforatum* constituents and the biological activity of the resulting postbiotic metabolites. This study aimed to evaluate the biotransformation of *H. perforatum* compounds by commercially available probiotic strains. Extract was obtained by triple extraction with 70% ethanol, concentrated under reduced pressure and lyophilized. Chemical composition of the extract and biotransformation of its compounds was assessed using UHPLC-DAD-MSn. Phytochemical analysis confirmed the presence of characteristic plant compounds, including rutin, hyperoside, isquercitrin, quercetin, hyperforin and adhyperforin. During incubation with most probiotics, rutin levels increased by more than 50% after 6h and decrease after 24h. Importantly, after 24h of incubation, a new postbiotic metabolite was observed in the presence of Dr. Max probiotic containing selected *Lactobacillus* and *Bifidobacterium* strains. New metabolite ($[M-H]^-$ m/z 245) produced MS² fragments at m/z 277, 201, 159, 125. These studies indicate a potential effect of biotransformation on the antidepressant activity of *H. perforatum* extract. Further studies will focus on the isolation and identification on the newly discovered metabolite.

Keywords: *Hypericum perforatum*, rutin, probiotics, UHPLC-DAD-MSn, postbiotic metabolite

P-43: Overcoming Microbiological Instability: Preservation Techniques for Spent Coffee Grounds as a Sustainable Substrate for Biotechnological Applications

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Spent coffee grounds (SCG) are a ubiquitous and nutrient-dense organic waste stream, characterised by a significant concentration of polysaccharides, nitrogen compounds, and essential minerals. In recent years, there has been a notable increase in the focus on SCG in the domain of applied microbiology, particularly with regard to its utilisation as a sustainable and cost-effective substrate for the cultivation of microorganism and edible fungi (Basidiomycota). A significant number of researchers have identified their potential and are endeavouring to process them into new, high-value products. Nevertheless, the suboptimal microbiological stability of this waste poses a significant challenge to its implementation, irrespective of the intended application. The overarching objective is therefore to identify scalable, and environmentally sustainable methods of microbiologically preserving them, ensuring sufficient quality for the intended application. The present study aims to compare traditional thermal methods of reducing microorganisms (sterilisation, pasteurisation and tyndallisation), with alternative methods, such as spontaneous fermentation under anaerobic conditions or exposure to elevated pH level. The study demonstrated that each of the analysed methods had a significant effect on the abundance of the microorganism population. Conventional thermal methods were found to be the most effective in completely reducing microflora. Nevertheless, alternative methods – such as fermentation and alkaline pH treatment – have been demonstrated to be efficacious in reducing undesirable microflora, whilst offering solutions that do not necessitate the use of specialised equipment.

Keywords: spent coffee grounds, fermentation, sterilisation, pasteurization, tyndallisation

P-44: Mechanism of action of new 4,5,6,7-tetrabromo-1H-benzimidazole derivatives in breast cancer

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Previous studies have demonstrated that 4,5,6,7-tetrabromo-1H-benzimidazole derivatives exhibit significant anticancer activity by targeting CK2, a key regulator of cellular processes related to proliferation and apoptosis. Its overexpression is noted in various cancers, including breast cancer, emphasizing its importance as a therapeutic target.

This research aimed to investigate the effects of two newly synthesized derivatives of 4,5,6,7-tetrabromo-1H-benzimidazole: 1-phenyl-3-(4,5,6,7-tetrabromo-2-methyl-1H-benzimidazol-1-yl)propan-1-one (**1a**), and 1-[(4,5,6,7-tetrabromo-1H-benzimidazol-2-yl)sulfanyl]propan-2-one (**2a**) against breast cancer. The effects of these new derivatives and their parent compounds, 4,5,6,7-tetrabromo-2-methyl-1H-benzimidazole (2Me-TBBI, **1**) and 4,5,6,7-tetrabromo-2-(methylsulfanyl)-1H-benzimidazole (K37, **2**), were analyzed on two cancer cell lines: MCF-7 (hormone-dependent breast cancer) and MDA-MB-231 (triple-negative breast cancer). Cytotoxicity, assessed via the MTT assay, showed enhanced anticancer activity of the derivatives. Cell proliferation was monitored through changes in confluence over time, revealing significant inhibition of cell growth in both lines. Further characterization through cell cycle analysis demonstrated cell arrest in specific phases. Additionally, immunodetection techniques assessed changes in the expression of proteins associated with CK2 kinase activity. The derivative **1a** exhibited the highest biological activity, significantly suppressing cell proliferation and effectively inhibiting the phosphorylation of anti-apoptotic proteins, leading to decreased cancer cell survival.

The findings suggest that structural modifications of 4,5,6,7-tetrabromo-1H-benzimidazole derivatives are an effective strategy to enhance anticancer activity and could support further research into new CK2 inhibitors.

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Keywords: breast cancer, CK2 protein kinase, CK2 protein kinase inhibitors, 4,5,6,7-tetrabromo-1H-benzimidazole

P-45: Cell line-dependent microRNA expression in canine osteosarcoma *in vitro* models

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Canine osteosarcoma (OSA) is an aggressive spontaneous malignancy and an important model in comparative oncology. OSA biology is shaped by multiple molecular mechanisms, including post-transcriptional regulatory pathways and microRNAs (miRNAs). The miR-17-5p, miR-15b-5p and miR-375-3p, have attracted attention as potential oncomiRs in extracellular vesicle-mediated intercellular communication. Because endogenous expression differs markedly among *in vitro* models, accurate baseline profiling is key. This study assessed the expression of selected oncomiRs in three canine OSA cell lines.

The analysis included three canine OSA cell lines, i.e. OSCA-8, OSCA-29 and D17 with reference to hTERT canine fibroblasts. Total RNA was isolated using the phenol–chloroform method, followed by polyadenylation and cDNA synthesis using the MiR-X miRNA First-Strand Synthesis Kit (Takara). Expression levels of selected miRNAs were then quantified by RT-qPCR. The obtained data were normalized using the RQMAX method. Statistical significance was assessed by one-way ANOVA.

RT-qPCR revealed distinct, cell line-dependent expression patterns. Specifically, miR-17-5p and miR-15b-5p exhibited downregulation within the OSCA29 model compared to the hTERT reference. In turn, miR-375-3p showed clear upregulation in OSCA8 and D17 lines, while remaining at baseline levels in OSCA29. All tested cancer lines displayed unique miRNA signatures, with OSCA29 emerging as the most molecularly distinct model. These findings confirm that canine OSA cell lines possess highly divergent non-coding RNA profiles.

Findings demonstrate molecular heterogeneity among canine OSA cell lines and indicate that miR-17-5p, miR-375-3p and miR-15b-5p expression may be useful for the biological characterisation of *in vitro* models. These results underscore the importance of careful model selection in studies on miRNA-related mechanisms and provide a baseline for future *in silico* target prediction.

Keywords: dog, bone tumor, non-coding RNA, gene expression, post-transcriptional regulatory pathways

P-46: Polysaccharide nanocarriers functionalized with targeting and chelating ligands for theranostics

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Despite significant advancements in modern medicine, cancer remains one of the leading causes of mortality worldwide. According to statistics acquired by World Health Organisation almost 30% of European population will develop cancer in their lifetime, with over 10% cases resulting in death. Conventional treatments, including immunotherapy and chemotherapy, often prove insufficient, and mortality rates remain high. Consequently, there is an increasing demand for research into alternative therapeutic approaches. This has led to the research on development of polysaccharide-based drug carriers, enabling efficient delivery and precise targeting while minimizing harm to healthy tissues. The aim of this study was to identify the most efficient method for functionalizing polymer nanomaterials based on carboxymethylcellulose with the chelating ligand DOTA and the CCZ02 peptide targeting receptors overexpressed on melanoma cells. For the experiment, coupling reactions were performed for each ligand with both the linear polymer and nanogels using 4-(4,6-dimethoxy-1,3,5-triazin-2-yl)-4-methylmorpholinium chloride as the coupling agent. Different reaction variants were applied, including systems supplemented with additional reagents such as 4-dimethylaminopyridine and N-methylmorpholine. The efficiency of carrier functionalization with the peptide was assessed using the BCA assay, a quantitative colorimetric method for peptide determination. The coupling efficiency of the chelating ligand to carboxymethylcellulose was evaluated by radiolabelling with ¹⁷⁷Lu. The results indicated that the efficiency depends on the ligand type, solvent used, and the presence of additives. Based on the obtained results, the most effective reaction system was selected, enabling high efficiency in the simultaneous coupling of a chelating ligand and a targeting peptide to nanocarriers.

Keywords: nanocarriers, chelating ligand, cancer, peptide

P-47: Effects of light intensity on the growth of selected duckweed species (*Lemna minor*, *Landoltia punctata*, and *Spirodela polyrhiza*)

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Duckweeds are fast-growing aquatic plants with high potential for biomass production and wastewater treatment. However, their growth responses to light intensity remain poorly understood. Therefore, this study evaluated the effect of light intensity within the photosynthetically active radiation range, expressed as photosynthetic photon flux density (PPFD), on the growth of three duckweed species: *L. minor*, *L. punctata*, and *S. polyrhiza*. In accordance with OECD 221 guidelines, plant growth was assessed after 7 days, using relative growth rate (RGR) based on frond number and frond surface area, with dry biomass included as an additional measure. In addition, light saturation points (I_{sat}) were determined for each parameter.

Across the tested range of 50-1220 $\mu\text{mol m}^{-2} \text{s}^{-1}$, *L. minor* showed the highest growth rates, whereas *L. punctata* and *S. polyrhiza* exhibited lower and relatively similar RGR values. All species showed an increase in growth between 50 and 390 $\mu\text{mol m}^{-2} \text{s}^{-1}$, but their responses differed at higher light intensities. *L. minor* continued to increase slightly up to the maximum tested intensity, *L. punctata* plateaued beyond 390 $\mu\text{mol m}^{-2} \text{s}^{-1}$, and *S. polyrhiza* reached maximum RGR at 610 $\mu\text{mol m}^{-2} \text{s}^{-1}$, followed by a slight decline at higher irradiance. Dry biomass also increased with light intensity and was highest at the maximum tested PPFD.

The calculated I_{sat} values for RGR based on frond number were 296 ± 35 , 286 ± 21 , and $160 \pm 17 \mu\text{mol m}^{-2} \text{s}^{-1}$ for *L. minor*, *L. punctata*, and *S. polyrhiza*, respectively. The corresponding I_{sat} values based on frond surface area were 274 ± 67 , 220 ± 7 , and $205 \pm 17 \mu\text{mol m}^{-2} \text{s}^{-1}$, while biomass-based I_{sat} values were higher, ranging from 343 ± 49 to $440 \pm 44 \mu\text{mol m}^{-2} \text{s}^{-1}$. These results indicate that increasing light dose above the saturation range does not proportionally enhance growth and highlight the importance of species-specific, energy efficient lighting strategies in controlled duckweed cultivation systems.

Keywords: *Lemna minor*, *Landoltia punctata*, *Spirodela polyrhiza*, light intensity, light saturation point

P-48: The effect of silver nanoparticles on *Toxoplasma gondii* tachyzoites

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Toxoplasma gondii is a cosmopolitan protozoan parasite that causes toxoplasmosis in humans, affecting 30–50% of the global population. While typically asymptomatic in healthy individuals, the infection may lead to severe complications in immunocompromised patients and congenitally infected infants. This is largely due to the parasite's ability to evade the host's immune system and cross the body's physiological barriers (e.g. blood-placenta and blood-brain barrier) to migrate to immunologically-privileged sites. The current problem in treating toxoplasmosis is a lack of a successful therapy that would also be relatively safe to use. At present, available treatment options are only partially effective and often associated with severe adverse effects, such as bone marrow suppression. Recent studies have demonstrated the antimicrobial potential of silver nanoparticles (AgNPs), including their enhanced efficiency when synthesised using tea extracts. The purpose of this study was to assess the potential effects that AgNPs and green tea extract could have on *T. gondii* tachyzoites. Parasites were incubated for 48h with the solutions of 1) AgNPs; 2) AgNPs synthesised using green tea extract (GT-AgNPs); and 3) green tea extract (GT); at concentrations of 1,25–10 $\mu\text{g}/\text{ml}$. After the 24h and 48h incubation periods, the parasite's cell count and viability were determined. The results showed that green tea extract alone did not significantly affect the parasite's survival. In contrast, the solution of AgNPs demonstrated the highest antiparasitic effect, reducing viability by 37–73% after 48h. The effect was both dose- and time-dependent, as higher concentrations and longer exposure led to greater reduction in the parasite's viability. Overall, the results suggest that AgNPs may serve as a promising alternative strategy for limiting *T. gondii* proliferation.

Keywords: Silver nanoparticles, Green tea, *Toxoplasma gondii*, Toxoplasmosis, Treatment

P-49: Structural and sequence determinants of efficient nonconventional splicing in *Euglena longa*: a comparison of constitutive and alternative introns

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Alternative splicing involves the excision of introns and the ligation of exons in various ways, resulting in different forms of mature mRNA being generated from a single pre-mRNA molecule. A remarkable feature of alternative splicing in euglenoids is the differentiated removal of nonconventional introns, which are not found in other organisms. These introns are characterised by a structure atypical for most eukaryotes, including the absence of classical consensus sequences at the ends (e.g., GT-AG). Crucially, they often maintain a conserved secondary structure through base pairing between the 5' and 3' splice site regions, potentially involving a CAG/CTG pairing element, which suggests unique mechanisms for their recognition and excision. Differentiated removal of nonconventional introns seems to be an important process in gene expression regulation in euglenids. The presented study focuses on comparing the conserved structural and sequence determinants between nonconventional introns that are constitutively and alternatively excised, in order to identify features that function as the primary splicing signals governing intron recognition efficiency. For this purpose, data on introns from selected genes were collected and compared. This made it possible to identify features that appear to be of crucial importance for successful intron recognition and excision. While no significant differences were observed at the ends of constitutive (CONS) and alternative (ALT) introns, ALT introns virtually lacked the critical CAG/CTG triplet, even though the base pairing structure itself remained preserved. This suggests that the presence or absence of the CAG/CTG pairing element acts as a major cis-regulatory determinant influencing efficient nonconventional introns removal in euglenoids.

Keywords: Alternative splicing, Non-conventional introns, *Euglena longa*, *Euglena*

P-50: Small heroes, great power: bacterial metabolites as sustainable alternatives for the farms of the future

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Agriculture is currently challenged by population growth, resource depletion, environmental degradation, and increasingly frequent extreme climatic events. Therefore, sustainable fertilization strategies that not only enhance crop efficiency but also reduce anthropogenic pressure on the environment are urgently needed.

In this context, a foliar micronutrient fertilizer based on bacterial metabolites, specifically siderophores and surfactin, was developed. These biodegradable compounds increase micronutrients bioavailability through metal chelation and improve their foliar uptake by reducing surface tension, potentially allowing for lower fertilizer input.

To preliminarily determine metal-binding efficiency and optimal ratios between components, complexation assays and siderophore (pyoverdine) fluorescence measurements were performed. Surface tension and contact angle analyses were used to identify the most effective surfactin concentration, while a seed germination assay was used to examine potential phytotoxic effects. In addition, a controlled phytotron experiment on basil *Ocimum basilicum* L. seedlings was conducted to preliminarily evaluate plant growth responses.

The results suggest a promising potential for further investigation and, in the long term perspective, may support development of a sustainable alternative to conventional micronutrient fertilizers, in line with the principles of sustainable agriculture and reduced-input systems.

Keywords: sustainability, foliar fertilizer, micronutrients, bacterial metabolites

P-51: Differentiation-Dependent Effects of ATP and Serotonin on Neuronal Cell Survival

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Purinergic and serotonergic signaling play key roles in regulating neuronal cell function, including survival, metabolism, and stress responses. Although the individual effects of ATP and serotonin (5-HT) have been widely studied, their combined impact on neuronal cells at different stages of differentiation remains unclear.

This study aimed to evaluate the effects of ATP, serotonin (5-HT), and their combinations on the viability and survival of SH-SY5Y cells in undifferentiated and neuronally differentiated states.

Experiments were conducted using the human neuroblastoma SH-SY5Y cell line. Differentiation was induced with retinoic acid (RA) and PMA. Cell viability was assessed using the MTT assay, while survival was evaluated with the Neutral Red Uptake (NRU) assay. Cells were treated with varying concentrations of ATP, 5-HT, and their combinations, and analyzed after 24 and 72 hours.

Both ATP and 5-HT showed concentration-dependent effects. High ATP levels reduced metabolic activity, particularly in undifferentiated cells. Serotonin displayed protective effects at low concentrations but became cytotoxic at higher doses. NRU results confirmed these findings, indicating changes in cell membrane integrity. Combined treatments produced variable effects depending on concentration and differentiation status, with serotonin modulating ATP-induced responses.

These findings indicate that ATP and serotonin exert complex, context-dependent effects on neuronal cells, influenced by differentiation status and concentration. Their interaction may be relevant for mechanisms of neuroprotection and neurodegeneration.

Keywords: Purinergic signaling, Serotonin (5-HT), ATP; SH-SY5Y cells, Neuronal differentiation;

P-52: Lineage Tracing Reveals the Contribution of MCAM-Positive Plastic Tumor Cells to Breast Cancer Progression

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Triple-negative breast cancer (TNBC) is a highly aggressive subtype with limited treatment options, presenting significant clinical challenges. Vascular mimicry (VM) describes the formation of vessel-like structures by cancer cells that function independently of angiogenesis. These structures support the delivery of nutrients and oxygen, promoting tumor growth and metastasis. We previously showed that MCAM-positive breast cancer cells are involved in VM. We hypothesize that these cells possess stem cell-like properties and can differentiate into endothelial-like cells, playing a key role in VM and metastatic spread. Understanding MCAM's role in tumor plasticity could lead to new therapeutic targets for TNBC. To explore MCAM's role in VM and cancer progression, we conducted a lineage-tracing analysis to determine whether endothelium-like MCAM-positive breast cancer cells are crucial for tumor vascular mimicry and invasion. We developed a lineage-tracing model in human breast cancer cell lines using a dual-plasmid system that includes an MCAM promoter-driven Cre-ER and an mTmG reporter. When MCAM-driven Cre-mediated recombination occurs, the mT-seq is excised, leading to permanent GFP labeling of MCAM-expressing cell progeny. These labeled cells were then injected into the fourth mammary gland of NOD-Scid mice. After tamoxifen induction following tumor formation, we observed that some GFP-positive tumor cells exhibited phenotypic plasticity, acquiring endothelial-like features and actively participating in VM. Additionally, we detected metastasis to the lungs.

In conclusion, these findings demonstrate that a subset of tumor cells with high plasticity can acquire an endothelial-like phenotype, as indicated by MCAM expression. This underscores the critical role of MCAM in tumor progression. Our study offers a novel perspective on breast cancer plasticity and suggests that targeting MCAM may reduce tumor aggressiveness and improve clinical outcomes.

Keywords: Triple-negative breast cancer, vascular mimicry, lineage-tracing, MCAM, tumor plasticity

P-53: Optimising mass of biochars from diverse feedstocks for humic substance recovery from two types of reject water

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Biochars (BCs) are recognized for their high sorption potential, with properties strongly influenced by the biomass feedstock and pyrolysis conditions. These characteristics make BCs widely used in agriculture as soil amendments, enhancing soil physicochemical properties and its capacity to immobilize contaminants. Recently, interest has grown in employing BCs to recover valuable organic compounds, such as humic substances (HSs), from reject waters generated during the centrifugation of anaerobically digested sludge in wastewater treatment plants. HSs are vital components of soil and water, and their conventional extraction relies primarily on non-renewable sources, making reject waters a promising alternative. This study evaluated the sorption potential of BCs for recovering HSs from two types of reject water: from a mono-fermentation system (RWGL) and a co-digestion system (RWKL). Five BCs derived from distinct biomass feedstocks: straw (S1 and S2), oak chips (O2), sunflower husks (F1), and digestate (D2), were analysed to assess their sorption properties and HS recovery efficiency. Optimal sorbent masses for each BC were determined, and the efficiency of HS recovery from reject waters was compared. For all BCs, the HS sorption capacity (q_e) decreased with increasing sorbent mass. For S1, q_e decreased from 68.31 to 3.33 $\text{mg}\cdot\text{g}^{-1}$ (for RWGL-derived HSs) and 109.61 to 6.49 $\text{mg}\cdot\text{g}^{-1}$ (for RWKL-derived HSs), while for S2, it decreased from 69.91 to 3.44 $\text{mg}\cdot\text{g}^{-1}$ (for RWGL-derived HSs) and 139.22 to 7.63 $\text{mg}\cdot\text{g}^{-1}$ (for RWKL-derived HSs). Similar trends were observed for O2 (44.53-2.73 and 127.46-4.71 $\text{mg}\cdot\text{g}^{-1}$), D2 (49.18-2.60 and 119.95-4.41 $\text{mg}\cdot\text{g}^{-1}$), and F1 (50.16-1.91 and 128.02-3.31 $\text{mg}\cdot\text{g}^{-1}$). The highest q_e values were consistently observed for RWKL-derived HSs, reflecting its higher HS content. These findings confirm the potential of BCs for HS recovery.

Keywords: Biochars, humic substances, recovery, reject waters, sorption

P-54: Design and Expression of Recombinant Mitochondrial Proteins in FreeStyleTM 293-F Cells.

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Mitochondria regulate essential cellular processes including energy metabolism, RNA homeostasis and programmed cell death through the coordinated action of multiple proteins. Among these, polynucleotide phosphorylase (PNPase), a conserved exoribonuclease, plays crucial roles in maintaining mitochondrial function. PNPase participates in RNA processing and degradation, however, it is possible that it may also be involved in additional aspects of riboregulation. Recent reports suggest its interaction with other proteins, like adenine nucleotide translocator 2 (ANT2), mediating the exchange of ADP and ATP across the inner mitochondrial membrane, and BAK, a proapoptotic protein involved in mitochondrial outer membrane permeabilization during apoptosis. Dysfunction of these proteins has been associated with mitochondrial disorders, development delay and neurological defects, therefore it is crucial to understand the molecular mechanisms underlying their functions and potential interplay. Elucidating PNPase, ANT2 and BAK functions requires access to the protein in a form closely resembling its native mitochondrial state in human cells, preserving proper folding, activity and organellar localization. In this study, recombinant constructs encoding PNPase, ANT2 and BAK fused to N- or C-terminal Strep-tag were designed to facilitate purification. The constructs were introduced into human cell line to enable expression within the physiological environment. Successful expression of recombinant protein was obtained, and the presence of the Strep-tag was confirmed using immunodetection methods. Mitochondrial localization was verified through analysis of subcellular fractions. Purification was established using affinity and size exclusion chromatography. The strategy provides a new approach for obtaining human proteins in a state resembling their native mitochondrial form and establishes a platform for future studies on their roles in mitochondrial function and human diseases.

Keywords: human PNPase, mitochondrial RNA metabolism, Strep-tag purification

P-55: AgNPs from ascorbic acid-rich plant waste: sonochemical synthesis and antimicrobial activity

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Due to the unique nanostructural properties of silver, there is a need to develop methods that enable precise control over nanoparticle nucleation and growth while minimizing the use of synthetic reducing agents. In this study, a sonochemical approach was employed for the synthesis of silver nanoparticles (AgNPs) using extracts derived from plant waste—parsley stems and potato peels—as a source of ascorbic acid and other redox-active compounds. Ascorbic acid acts as a reducing agent, converting Ag^+ to Ag^0 , while ultrasound, through acoustic cavitation, generates localized high-temperature and high-pressure conditions, enhancing reaction kinetics and influencing nucleation and growth processes.

The obtained colloidal systems differed in stability, dispersity, and particle size distribution depending on the composition of the plant extract and sonochemical parameters, indicating the significant role of both biomass composition and process conditions in shaping AgNP properties. The presence of organic compounds in the extracts suggests their additional role as capping agents, contributing to nanoparticle stabilization and preventing aggregation.

Antimicrobial activity was evaluated using agar overgrowth assays (semi-quantitative scale) and disc diffusion tests against Gram-positive bacteria (*Staphylococcus aureus*), Gram-negative bacteria (*Escherichia coli*), and yeasts (*Candida albicans*, *Pfaffia rhodozyma*, *Yarrowia lipolytica*, *Saccharomyces cerevisiae*). The effectiveness of AgNPs depended on their physicochemical characteristics, concentration and incubation time. Yeasts, particularly *P. rhodozyma* and *C. albicans*, exhibited the highest sensitivity, whereas Gram-negative bacteria (*E. coli*) showed higher resistance.

These results demonstrate that combining sonochemistry with naturally occurring reducing agents present in plant waste enables controlled synthesis of AgNPs with antimicrobial properties, while aligning the process with the principles of green chemistry.

Keywords: silver nanoparticles, AgNPs, sonochemical synthesis, antimicrobial activity, plant waste

P-56: Biotechnological Potential of Fabaceae *In Vitro* Cultures for Phenolic Compound Production with Cosmetic Applications

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Phenolic compounds represent a diverse group of plant secondary metabolites involved in defense against environmental stressors, such as UV radiation and pathogens, and are widely recognized for their antioxidant, anti-inflammatory, and photoprotective properties relevant to cosmetic applications. However, the identification of efficient, sustainable, and controllable sources of these bioactive compounds remains a significant challenge. *In vitro* cultures offer a controlled platform for enhancing their production through the application of biotic or abiotic elicitors.

The aim of this study was to evaluate the potential of selected *Fabaceae* species, such as *Medicago sativa*, *Medicago truncatula*, and *Pisum sativum* as sources of phenolic compounds. *Fabaceae* plants, often overlooked in cosmetology and primarily associated with food or feed production, may serve as valuable sources of bioactive metabolites, although their cosmetic potential remains poorly explored.

Leaf tissues and callus cultures were analyzed for total phenolic content (TPC) and antioxidant activity (FRAP). The results showed significant differences between species and tissue types. The highest phenolic content was observed in *M. truncatula*, with callus cultures exhibiting levels comparable to leaves and approximately twofold higher than in the other species, while *M. sativa* and *P. sativum* showed lower accumulation. Antioxidant activity followed a similar trend, with the highest values in *M. truncatula* leaves and lower activity in callus cultures; a positive correlation between phenolic content and antioxidant capacity was observed.

These findings indicate that both species selection and tissue differentiation significantly influence phenolic biosynthesis, with *M. truncatula* callus cultures representing a promising and sustainable platform for the production of bioactive compounds.

Keywords: Fabaceae, *In vitro*, Cosmetics, Phenolic Compounds, Antioxidants

P-57: Targeting replication stress: MK-8776 and disulfiram trigger apoptosis in ovarian cancer with cell line-specific kinetics

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Ovarian cancer remains one of the most lethal gynecological malignancies, largely due to late diagnosis and the development of resistance to conventional therapies. Targeting mechanisms that increase replication stress and promote apoptotic cell death has emerged as a promising therapeutic strategy.

To better recapitulate tumor-like conditions, cells were cultured as 3D spheroids, which more accurately model microenvironmental factors such as cell-cell and extracellular matrix interactions, nutrient gradients, and drug penetration compared with conventional 2D cultures. Cytotoxicity was measured using the resazurin reduction assay. Drug concentrations for subsequent analyses were selected based on preliminary combination effects and were set at 5 μM MK-8776 with 2.5 μM disulfiram for TOV-21G and 2.5 μM MK-8776 with 5 μM disulfiram for OVCAR-8.

The aim of this study was to evaluate the apoptotic response induced by the combination of the CHK1 inhibitor MK-8776 and disulfiram in ovarian cancer cell lines. Apoptosis was assessed by measuring caspase-3/7 activity using flow cytometry after 48 h of treatment and by Western blot analysis of PARP-1 cleavage and caspase-3 expression after 48 h and 120 h.

Flow cytometry revealed an increase in the percentage of caspase-3/7-positive cells in TOV-21G cells after 48 h. In contrast, OVCAR-8 cells showed only a modest increase in caspase-positive cells, although the median fluorescence intensity of the caspase-3/7 signal increased in the combination group. Western blot analysis demonstrated different temporal patterns of apoptosis between the two cell lines. In OVCAR-8 cells, PARP-1 cleavage was detected after 48 h and remained at 120 h, whereas in TOV-21G cells cleaved PARP-1 appeared only after prolonged treatment (120 h). These findings indicate that MK-8776 combined with disulfiram induces apoptotic signaling in ovarian cancer cells with cell line-specific response kinetics.

Keywords: Ovarian cancer, CHK1 inhibition, Disulfiram, apoptosis

P-58: *In silico* analysis of transcription factor binding sites of plastoglobule-related genes in response to abiotic stress in the *Arabidopsis thaliana* model

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The aim of *in silico* analysis was to identify transcription factor binding sites (TFBSs) in a set of plastoglobule-related genes and to confirm whether the transcription factors (TFs) binding to those sites participate in abiotic stress responses in *Arabidopsis thaliana*. Plastoglobules are plastid-localized lipoprotein particles attached to the stromal side of thylakoid membranes. The analysis was performed on a set of 32 plastoglobule-related gene IDs, including that of *VTE1* encoding tocopherol cyclase, a plastoglobule-localized enzyme catalyzing the biosynthesis of α -tocopherol, the most biologically active form of vitamin E. In the TFBS Discovery Tool Hub (TDTHub), the Find Individual Motif Occurrences (FIMO) algorithm significantly associated 17 genes of the set, including *VTE1*, with a TFBS motif most likely bound by Promoter Binding Factor 3 (DPBF3), a basic leucine zipper (bZIP) TF. The frequency matrix alignment in a JASPAR tool showed DPBF3's significant similarity in its DNA binding site to Long Hypocotyl 5 (HY5), a light-mediated bZIP TF involved in cold acclimation. Furthermore, the promoter analysis of *VTE1* in PlantPAN 4.0 suggests that the expression of *VTE1* is regulated by TFs involved in the positive regulation of cold acclimation and drought tolerance. Multiple bZIP TFBSs, including the ones recognized by HY5 and abscisic acid (ABA)-inducible TFs, were found *in silico* in *VTE1*'s promoter. Co-expression analysis results suggest that *VTE1* is co-expressed with TFs involved in cold-inducible processes, such as flowering. Additionally, chromatin immunoprecipitation sequencing (ChIP-seq) data from PCBase 2.0 suggest preferential binding of TFs to *VTE1*'s promoter under dark conditions and with ABA treatment, when plants generally have higher drought tolerance. It has been concluded that the analyzed genes are regulated by environmental conditions and that this further confirms the crucial role of plastoglobules in abiotic stress responses in *A. thaliana*.

Keywords: *in silico* analysis, plastoglobules, abiotic stress responses, transcription factor binding sites, *Arabidopsis thaliana*

P-59: Combined Surface-Enhanced Raman Spectroscopy And Chemometric Analysis Of Blood Plasma And Bone Marrow For Rapid Detection And Monitoring Of Pediatric Acute Lymphoblastic Leukemia

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Acute lymphoblastic leukemia (ALL) is the most prevalent pediatric malignancy and requires rapid, accurate, and minimally invasive diagnostic strategies. In this study, we investigate the applicability of surface-enhanced Raman spectroscopy (SERS) combined with chemometric analysis for label-free detection and monitoring of pediatric ALL using clinically relevant biofluids. SERS spectra were collected from blood plasma and bone marrow samples obtained from pediatric patients at diagnosis, during induction and consolidation therapy, and from healthy controls. Multivariate statistical methods, including partial least squares regression, enabled effective discrimination between disease states and treatment stages, demonstrating high sensitivity to subtle biochemical variations.

Distinct spectral features at 724, 1002, 1394, and 1450 cm^{-1} were identified as key biomarkers associated with nucleic acids, proteins, and lipid metabolism. The analysis revealed decreased phenylalanine levels in bone marrow and significant alterations in purine-related metabolites in plasma, reflecting leukemia-associated metabolic reprogramming. Moreover, convergence of spectral profiles during treatment indicates a measurable biochemical response to therapy.

The results demonstrate that SERS enables rapid, non-invasive molecular characterization of leukemia, capturing both systemic and local biochemical changes with minimal sample preparation. This approach holds strong potential for early diagnosis, treatment monitoring, and advancement of precision medicine in pediatric oncology.

Keywords: surface-enhanced Raman spectroscopy, pediatric acute lymphoblastic leukemia, chemometric analysis, partial least squares regression, liquid biopsy

P-60: Green synthesis of gold nanoparticles using milk thistle seed extract and their cytotoxicity toward skin cells

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Nanoparticles, due to their unique physicochemical and biological properties, are finding increasingly wide applications, including medicine, pharmacy and environmental protection. Among them, gold nanoparticles have received significant interest and are used in skincare cosmetics due to their anti-aging, protective and proregenerative effects. The most commonly used chemical and physical synthesis methods, despite their high efficiency and the ability to control process parameters, are associated with limitations, such as the generation of toxic waste and environmental pollution. Green synthesis has emerged as an attractive alternative. This method uses natural materials, such as plant extracts, containing secondary metabolites capable of reducing metal ions and stabilizing newly formed nanoparticles. Milk thistle (*Silybum marianum*) seed extract, traditionally used for liver disorders, contains silymarin, which is a compound with strong antioxidant, anti-inflammatory, and immunomodulatory properties. These features can potentially enhance the positive effects of gold nanoparticles on the skin. The aim of the study was to assess the cytotoxicity of gold nanoparticles obtained by green synthesis using silymarin toward skin cells. Gold nanoparticles were produced using milk thistle seed extract and silymarin from a medicine. The physicochemical properties of the nanoparticles, including their size, polydispersity and morphology, were characterized using electron microscopy, dynamic light scattering (DLS) and nanoparticle tracking analysis (NTA). UV-vis spectra were also recorded. To evaluate cytotoxicity, HaCaT cells were treated with various concentrations of the synthesized nanoparticles. Cell morphology was examined using the May-Grünwald-Giemsa staining method, while metabolic activity was assessed with the XTT assay. The results confirmed the successful green synthesis of gold nanoparticles and their biocompatibility within the tested concentration range.

Keywords: gold nanoparticle, green synthesis, silymarin, milk thistle

P-61: Affinity of plant Rab proteins for REP protein

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Rab proteins belong to the superfamily of small GTPases. They are responsible for regulating intracellular vesicular transport, i.e., the formation, movement, docking, and fusion of vesicles. Their activity depends on posttranslational prenylation—geranylgeranylation—which enables membrane fusion. Rab escort protein (REP) plays a key role in this process, binding newly synthesized Rab proteins, presenting them to geranylgeranyl transferase, and facilitating their transport to target membranes. Rab–REP interactions are therefore essential for proper Rab maturation and cellular transport.

The aim of this study was to determine the affinity of selected plant Rab proteins for REP protein. Genes encoding selected Rab and REP proteins were cloned into appropriate expression vectors, and heterologous overexpression was performed in *Escherichia coli* using IPTG (isopropyl- β -D-1-thiogalactopyranoside). The obtained proteins were purified by affinity chromatography to obtain samples suitable for biophysical analysis. Rab–REP interactions were studied using microscale thermophoresis (MST), a sensitive technique that allows for the quantitative analysis of biomolecule binding in solution with minimal sample consumption. By monitoring changes in the thermophoretic mobility of fluorescently labeled REP in the presence of increasing Rab concentrations, dissociation constants were determined.

Comparing the dissociation constants of Rab proteins from different subfamilies to the REP protein will allow us to determine which of them are preferentially geranylgeranylated in plant cells. In the future, this knowledge may be used to construct mutants in the genes encoding individual Rab proteins to obtain plants with improved nutritional properties and increased resistance to pathogens.

Keywords: Rab proteins, REP, affinity, microscale thermophoresis

P-62: Selection and characterization of transgenic *Arabidopsis thaliana* plants with knocked out gene encoding the mitoribosomal protein Rps23

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Plant mitochondria play an important role not only in energy metabolism, but also in the regulation of genes and responses to environmental stress. A large part of these functions is due to the involvement of mitochondrial ribosomes (mitoribosomes), which influence the course of translation and, consequently, the synthesis of mitochondrial proteins.

During the experiment, the function of the nuclear-encoded protein Rps23, a component of the small subunit of the mitoribosome in *Arabidopsis thaliana* was analyzed in the context of its importance for genomic integrity and cellular function.

The aim was to obtain homozygous lines with the expression of the Rps23 protein knocked out using T-DNA insertion mutants. Zygosity was assessed based on selection on media containing sulfadiazine and PCR analyses using primer specific for the gene and the insertion. No homozygous individuals were obtained, which indicates the lethality of the mutation and suggests a key role of Rps23 in maintaining basic cellular processes.

Phenotypic analysis of heterozygous lines conducted over 6 weeks under long-day conditions did not show statistically significant differences compared to the wild type. Expression analysis using RT-qPCR showed a comparable level in both types of plants. At the same time, uniform resistance of the progeny to the antibiotic suggests the presence of additional copies of the resistance gene outside the insertion site.

Obtained results indicate that RPS23 is a gene essential for plant survival, and its partial knockout does not significantly affect the phenotype under standard conditions.

Keywords: *Arabidopsis thaliana*, plant mitochondria, mitochondrial ribosomes, Rps23 protein, gene expression

P-63: Event-Based Laser Speckle Imaging for Real-Time Assessment of Microbial Activity Across Varying Inoculum Densities

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Real-time monitoring of microbial activity is essential for advancing phenotypic characterization in microbiology. Traditional methods often lack the temporal resolution to capture early-stage dynamics, particularly when comparing cultures inoculated at different optical densities (OD). This study introduces event-based laser speckle imaging (LSI), combined with an event-based camera, as a novel approach to quantify microbial activity through speckle event rates.

Using *Saccharomyces cerevisiae* and *Escherichia coli* as model organisms, macrocolonies were inoculated from cultures at initial OD values of 1, 5, 10, and 50. The results demonstrate that event-based LSI can sensitively detect variations in speckle activity, with higher starting ODs yielding faster observable increases in speckle counts. This relationship highlights the method's potential to rapidly assess microbial growth dynamics based on inoculum density.

The integration of an event-based camera with LSI provides high temporal resolution and minimal data redundancy, enabling precise, label-free monitoring of microbial activity. By quantifying speckle event rates, this approach offers a robust tool for real-time analysis of microbial behavior. Future applications may extend to optimizing culture conditions, accelerating phenotypic screening, and enhancing antimicrobial susceptibility testing. This proof-of-concept study underscores the value of event-based LSI in providing dynamic insights into microbial activity, addressing a key need for faster and more informative microbiological assays.

Keywords: Laser speckle imaging, microbial activity, inoculum density, real-time monitoring

P-64: *Aralia spinosa* L. hairy roots as a biotechnological platform for araloside A production: optimization of *in vitro* culture conditions, growth kinetics, and biosynthetic efficiency assessment

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Hairy roots generated by *Rhizobium rhizogenes*-mediated transformation are attractive *in vitro* platforms for plant biotechnology due to rapid growth, genetic stability, high biosynthetic potential, and suitability for sustainable, environment-independent production of valuable plant metabolites without plant growth regulators.

In this study, the AS.T-J1 hairy root line of *Aralia spinosa* L., obtained using *R. rhizogenes* strain A4 and molecularly verified, was used as a model system for the optimization of araloside A, a triterpenoid saponin. The effects of medium composition, sucrose concentration, pH, and inoculum density on biomass growth and araloside A biosynthesis were optimized in flask cultures, and methanolic extracts were analyzed by UHPLC-DAD-ESI-MS3.

The culture productivity was additionally assessed using biosynthetic efficiency per inoculum index (BEI), an integrative parameter proposed by our group, expressed as metabolite content multiplied by dry harvest biomass and divided by inoculum fresh weight. This parameter reflects the amount of metabolite produced per gram of starting inoculum and combines biomass increase with metabolite accumulation.

Among the tested variants, Schenk and Hildebrandt medium (1972) supplemented with 50 g/L sucrose, pH 5.8, and 1.0% inoculum provided the best balance between biomass growth and araloside A biosynthesis. In a 90-day culture experiment, the growth curve was established at 5-day intervals, enabling the identification of distinct growth phases and determination of the optimal harvest time. BEI for araloside A reached its maximum value on day 45, indicating the highest production efficiency, whereas visible signs of culture aging appeared after day 75.

These findings demonstrate that *A. spinosa* hairy roots constitute a promising biotechnological platform for araloside A production and that BEI is a useful parameter for selecting optimal culture conditions and harvest time in *in vitro* systems.

Keywords: hairy roots, *Aralia spinosa*, biosynthetic efficiency index, araloside A, culture optimization

P-65: Plant responses to persistent pollutants: bioaccumulation and stress effects of perfluoroalkyl substances in *Solanum dulcamara in vitro*

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Perfluoroalkyl substances (PFASs) are chemicals that contaminate various environments. Due to their chemical properties which make them resistant for degradation, they are considered “forever chemicals”. PFAS can enter the environment through industrial processes and improper waste disposal. There are multiple studies on their negative impact on human health and ecosystems. One of the main sources of PFAS for humans is food consumption, for example by contaminated crops. *Solanum dulcamara* is a plant which is a representative of the *Solanaceae* family, to which belong many agricultural species like potato, tomato and eggplant. This study evaluates *S. dulcamara*'s response to one of the most commonly detected PFAS in the soil. The experiment was based on the *in vitro* cultivation of plants exposed to varying concentrations of tested substance. Focusing on biochemical stress markers and LC-MS/MS analysis of bioaccumulation in different plant tissues, this study provides new insights that may have an impact on strategies aimed at the remediation of contaminated agricultural lands and deeper understanding of the plant's defense mechanisms in the context of exposure to abiotic stress.

Keywords: PFAS, abiotic stress, bioaccumulation, plant tissue, Solanaceae

P-66: Enhancing the Specificity of DNA Methylation Monitoring through Effective Removal of Non-Specifically Adsorbed HRP: A Comparative Approach

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As a clinically relevant biomarker, DNA methylation, particularly 5-methyl cytosine has been investigated for the accurate diagnosis and treatment of various diseases, including cancer. However, since it occurs at extremely low concentrations in physiological fluids, its precise determination in the presence of interfering species remains challenging. Furthermore, the elimination of non-specifically adsorbed (NSB) proteins is crucial, as they are indistinguishable from the true signal and can therefore mislead the analysis.

In this study, we developed an electrochemical DNA hybridization assay for sensitive methylation detection, with particular emphasis on the elimination of NSB of HRP-IgG involved in the signal-generation mechanism. The effect of various surface blocking proteins such as BSA, casein and gelatin on DNA probes of varying chain lengths was performed to determine their influence on assay selectivity and sensitivity.

This work begins with initial immunoassay verification, followed by electrochemical assessment, and culminates in performance evaluation under microfluidic conditions using SPR measurements. Together, these results highlight the system's potential for the future development of microfluidic sensors for ultrasensitive detection of methylated DNA.

Keywords: DNA methylation, Non specific adsorption, blocking proteins, amperometry

P-67: Comparative Genomic Analysis and Experimental Validation of Genetic Variants in Five Cucumber (*Cucumis sativus* L.) Lines

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As a member of the Cucurbitaceae family, the cucumber (*Cucumis sativus* L.) is a globally significant horticultural crop, valued for its nutritional properties and versatility. In 2023, global production reached nearly 98 million tons. Its short life cycle, ease of cultivation under controlled conditions, and well-characterized genome make the cucumber a useful model for advancing biotechnological and genomic studies.

Cucumber reproductive biology is complex, with phenotypes including monoecious, gynoecious, and andromonoecious forms. Sex determination depends on genetic, hormonal, and environmental factors, and is fundamentally tied to ethylene-mediated flower differentiation, offering insights for plant biology and breeding.

Exploiting the natural genetic diversity within the species is crucial for agricultural initiatives focused on boosting yields, building disease resistance, and enhancing stress tolerance. Specific genetic mutations - namely single nucleotide polymorphisms (SNPs), insertions, and deletions - can profoundly alter the morphological, metabolic, and physiological traits that dictate a plant's resilience. Understanding these genetic variations is essential for the precise selection of superior genotypes and the refinement of farming strategies.

In this study, we conducted a comparative genomic analysis of five distinct cucumber breeding lines (2gg, 2667, 859, Gy3, and Hgy3) relative to the B10 reference genome. Identified genetic variations, including SNPs and indels, were validated using PCR and Sanger sequencing. The results confirm computational predictions and support the development of molecular markers for modern breeding, contributing to sustainable agriculture and food security.

This research was funded by a project from the National Science Center UMO2020/37/B/NZ9/00586

Keywords: *Cucumis sativus*, genetic variants, Sanger sequencing, B10 line

P-68: Plant-based expression systems: A novel platform for producing recombinant therapeutic proteins

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Introduction:

Recombinant therapeutic proteins are essential in modern oncology and immunology. Due to the high costs of mammalian systems and limitations of bacterial platforms, there is a demand for viable alternatives. Plant-based expression systems offer cost-effectiveness, scalability, absence of animal-derived components, and easy adaptation to GMP requirements, making them a competitive solution for protein production.

Aim of the study:

To develop a novel plant expression system based on *in-vitro* suspension cultures of *Nicotiana tabacum* BY-2 cells for the production of recombinant proteins.

Methods:

A plasmid carrying an expression cassette under the constitutive CaMV 35S promoter was synthesized using the pCAMBIA1305.1 vector. To validate the system, two proteins were investigated: (i) green fluorescent protein (GFP) as a reporter and (ii) fibroblast growth factor (FGF) as a therapeutic candidate. Plasmids were introduced into *Agrobacterium tumefaciens* LBA4404 via electroporation. The bacteria were used to agroinfect BY-2 cells, which were subsequently cultured for 4 weeks on modified LS medium supplemented with hygromycin B and cefotaxime. Transformation was confirmed via fluorescence microscopy and Western blot analysis.

Results:

A stably transformed BY-2 cell line producing GFP was successfully established. Fluorescence microscopy confirmed the presence of the product within plant cells and intracellular vesicles.

Conclusions:

The study demonstrates an effective plant-based method for recombinant protein production, enabling stable synthesis in the *N. tabacum* BY-2 line within 28 days with minimal effort and cost.

Keywords: expression system; agroinfection; recombinant protein

P-69: IFIT1 oligomerization kinetic study

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The innate immune response is the first line of defense against pathogens and relies on coordinated mechanisms involving specialized cells and proteins that detect and eliminate foreign molecules. Among them, IFIT family proteins play a key role in recognizing viral nucleic acids and limiting viral replication. This study focuses on IFIT1, which binds viral RNA containing a 5'-triphosphate group (ppp-RNA) or a cap 0 structure, and examines its ability to form oligomeric assemblies and the kinetic of this process. To achieve this, the native IFIT1 protein and its mutants, in which the domain responsible for homodimerization was altered, were expressed. Dynamic Light Scattering (DLS) was used for initial structural characterization, while oligomerization kinetic were assessed using thioflavin T fluorescence, including the effects of ATP and RNA.

DLS analysis revealed that native IFIT1 and the single mutant formed predominantly dimeric and octamer-like species at 15°C and 25°C, but underwent extensive aggregation into large oligomers at 37°C. In contrast, the double mutant displayed only one oligomeric form at each temperature, shifting from dimers at lower temperatures to large oligomers at 37°C.

Kinetic studies showed that IFIT1 WT alone exhibited a phase of active oligomerization followed by a saturation plateau. Increasing ATP concentrations reduced both the rate of oligomerization and the plateau level. Short RNA (~200 nt) had a similar inhibitory effect. In contrast, long RNA (~1800 nt) accelerated the oligomerization phase but resulted in a lower plateau, followed by a gradual decline in signal over time.

Overall, this results indicate that long RNA facilitates rapid formation of transient oligomeric species but does not promote their stable accumulation, whereas short RNA mainly inhibits oligomer formation in a manner similar to ATP. These findings provide new insight into IFIT1 behavior and its role in the innate immune response.

Keywords: immune response, IFIT proteins, IFIT1, viral infections, protein oligomerization

P-70: Comparison of the effect of fetal bovine serum (FBS) and bovine calf serum (BCS) on the development of ferroptosis resistance in the HT-1080 fibrosarcoma cell line

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Ferroptosis is a form of regulated cell death, defined by the accumulation of reactive oxygen species that cause lipid peroxidation and rupture of the cell membrane. Exploiting this mechanism shows promise for cancer therapy. The tumor microenvironment may influence cellular susceptibility to ferroptosis. *In vitro*, the microenvironment is represented by the culture medium and serum. Due to different developmental stages, serum obtained from bovine fetuses and calves may distinctly affect experimental outcomes. This study aimed to compare the effects of fetal bovine serum (FBS) and bovine calf serum (BCS) on ferroptosis resistance in the HT-1080 fibrosarcoma cell line. Cells were cultured in medium with either FBS or BCS, then exposed to increasing concentrations of the ferroptosis inducer ML162. Ferrostatin-1 was administered to a subset of cells to suppress ferroptosis. ML162 cytotoxicity was assessed using the CCK-8 assay and plate reader; and lipid peroxidation was evaluated using the C11-BODIPY probe, flow cytometry, and confocal microscopy. Cells grown in BCS-supplemented medium were more resistant to ferroptosis than those in FBS-supplemented medium, suggesting the presence of ferroptosis-inhibiting components in calf serum. These findings further underscore the importance of studying ferroptosis under varied culture conditions to better understand this cell death mechanism within the context of the human tumor microenvironment.

Keywords: ferroptosis, cell culture sera, fibrosarcoma, tumor microenvironment

P-71: C-methylated polytrimethylenimines: synthesis, antimicrobial properties, and the role of molecular mass

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The growing problem of antibiotic resistance highlights the need for new antimicrobial compounds with alternative mechanisms of action[1]. Cationic polymers, such as polyamines (e.g., linear polyethylenimine – L-PEI or linear polytrimethylenimine – L-PTMI), have emerged as a promising class of agents, as they can disrupt bacterial membranes, leading to rapid cell death while minimizing the risk of resistance development[2,3].

This study aims to investigate the effect of polymer molecular mass on its antimicrobial activity and cytotoxicity. Herein, we present the synthesis of linear polytrimethylenimine (L-PTMI) derivatives bearing alkyl groups incorporated into the polymer backbone with varying molecular mass. The materials were obtained via the cationic ring-opening polymerization (CROP) of substituted 2-oxazine monomers and subsequent acidic hydrolysis of poly(2-alkyl-2-oxazines). The antimicrobial activity of the synthesized polymers was evaluated by determining the minimum inhibitory concentration (MIC) against *Escherichia coli*, *Staphylococcus aureus* and *Candida albicans*. Furthermore, cytotoxicity was assessed using the MTT assay to establish dose-response curves and calculate IC₅₀ values for each compound across selected human skin (A375, HaCaT) and lung (A549, MRC-5) cell lines. The synthesized polymers represent a step toward the development of new antimicrobial materials with tunable properties and reduced risk of resistance development.

Keywords: antimicrobial agents, polymers, linear polytrimethylenimine, toxicity

P-72: The impact of JAO2 gene mutation on secondary growth and xylem formation in the *Arabidopsis thaliana* stem: anatomical and gene expression analyses

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Jasmonic acid oxidases (JAOs) represent an important group of enzymes responsible for the conversion of jasmonic acid (JA) into its inactive form, 12-hydroxyjasmonic acid (12OH-JA). Disruptions in the expression of genes encoding JAOs can lead to the accumulation of JA, which affects numerous developmental processes in plants as well as their defense responses to environmental stresses. Among these processes is xylogenesis - a highly coordinated developmental process including cambial cell division, cell expansion, secondary cell wall (SCW) deposition, lignification, and ultimately programmed cell death. Although secondary xylem formation is crucial for both ecosystems and the global economy, the molecular mechanisms governing SCW formation remain incompletely understood.

Given the results of previous studies indicating the significant role of the JAO2 protein in maintaining proper jasmonic acid levels in *Arabidopsis thaliana* tissues, our objective was to determine the impact of mutations in the *JAO2* gene on processes related to secondary growth, particularly in the context of xylogenesis (wood formation).

Anatomical analysis of the stems of *jao2* mutants compared to the wild type (WT) revealed significant changes, including an increased proportion of secondary xylem and a higher number of cambial cells. Furthermore, gene expression analyses indicated that the mutation in the *JAO2* gene leads to significant alterations in the expression levels of genes involved in secondary cell wall synthesis, including those related to lignin and other cell wall components. Together, these results highlight the crucial role of the *JAO2* gene and jasmonate catabolism in modulating secondary cell wall formation and controlling the development of vascular tissues.

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Keywords: JAO, xylogenesis, *Arabidopsis thaliana*, secondary growth of the stem, anatomy

P-73: Regulating MMP-9-Dependent Neuronal Plasticity In Situ

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Fragile X syndrome (FXS) is the most common monogenic cause of autism spectrum disorder-like symptoms and is associated with impairments in synaptic plasticity, cognitive function, and social behavior. Numerous studies indicate that increased activity of matrix metalloproteinase-9 (MMP-9) represents one of the key mechanisms underlying these deficits. A potential therapeutic strategy may therefore involve the use of TIMP-1, a natural inhibitor of this enzyme.

The aim of this project is to develop and analyze methods for regulating MMP-9-dependent plasticity in situ in an *Fmr1* knockout mouse model. The study seeks to determine whether intravenous delivery of TIMP-1 using nanoparticles can cross the blood–brain barrier and influence neural circuits underlying social behavior, with particular emphasis on the medial prefrontal cortex (mPFC).

Current work is focused on the production of TIMP-1 protein. In subsequent stages, TIMP-1 will be delivered using specialized nanoparticles. The effects of the intervention on social behavior will be assessed using the Eco-HAB system, enabling automated and objective monitoring of social interactions in group-housed mice under ecologically relevant conditions.

As the project is still at an early stage, the presented work focuses on the scientific rationale, experimental design, and optimization of the protein delivery system. The results of this study may contribute to the development of minimally invasive strategies for modulating MMP-9 activity, which is of particular relevance in the context of neuropsychiatric disorders associated with impaired brain plasticity.

Keywords: Fragile X syndrome, synaptic plasticity, social behavior, MMP-9, TIMP-1

P-74: Selenium biofortification of Chinese cabbage using a native plant growth-promoting bacterial consortium

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Selenium (Se) is an essential micronutrient; however, its low concentration in soils in many regions, including Poland, contributes to dietary deficiencies in humans and animals. Inorganic Se fertilization can mitigate this deficiency, but excessive application poses environmental risks. The combined use of beneficial microorganisms and low Se doses represents a promising strategy for producing Se-enriched functional foods. This study aimed to isolate native *Bacillus* and *Pseudomonas* strains from agricultural soils and evaluate their capacity to enhance Se accumulation in Chinese cabbage (*Brassica rapa L. subsp. pekinensis*). Native strains were isolated from agricultural soils of the Masovian Voivodeship using the heat-shock method (*Bacillus spp.*) and King's B medium (*Pseudomonas spp.*). Isolates were screened for plant growth-promoting traits, including phosphate and potassium solubilization, siderophore production, and indole-3-acetic acid (IAA) synthesis. Taxonomic identification was performed by 16S rRNA gene sequencing. Baseline soil Se was quantified by ICP-MS. Based on their plant growth-promoting traits, three strains representing *Bacillus* and *Pseudomonas* were selected to form a native consortium. Baseline soil Se concentration was below the limit of quantification. Greenhouse experiments will evaluate, assessing Se accumulation and speciation, plant biomass, and soil enzymatic activity.

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Keywords: Functional food, selenium biofortification, plant growth-promoting bacteria, *Bacillus*, *Pseudomonas*, Chinese cabbage, native bacterial consortium

P-75: Long-read metabarcoding and single-cell genomics: uncovering protist diversity in Masurian peatlands

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Protists (unicellular microbial eukaryotes) represent most eukaryotic lineages and inhabit all terrestrial and aquatic environments. Their typically small size and limited morphological features have historically constrained studies of their diversity, and cultivation difficulties have further hampered biological and ecological research. Advances in molecular methods, especially Next-Generation Sequencing (NGS), now overcome these limits. Environmental DNA metabarcoding and culture-independent genomic and transcriptomic analyses reveal protists as highly diverse and major ecosystem components. Still, research remains uneven, with most studies focusing on marine and large freshwater systems, while peatbogs remain underexplored.

This study compared culture-independent methods for characterizing protist diversity in the Masurian Lake District peatlands, Poland. First, long-read metabarcoding identified the most diverse and abundant taxonomic groups. Next, a selected taxon of heterotrophic euglenids was analysed using single-cell genomics. Finally, consistency between both methods in detecting protist taxa was assessed.

Long-read metabarcoding revealed that Alveolata, particularly apicomplexan gregarines, are widespread across peat bog sites. Euglenozoa, especially the genus *Petalomonas*, were frequent in the studied region. The distribution of *Petalomonas sphagnophila* matches its preference for acidic, oligotrophic environments associated with *Sphagnum* mosses. Single-cell isolation and genome amplification enabled molecular characterization and confirmed phylogenetic placement of the environmental sequences.

Overall, combining metabarcoding with single-cell genomics provided a comprehensive approach to analysing protist diversity in peatlands. The findings highlight the ecological role of key taxa and improve understanding of microbial eukaryotes in understudied ecosystems, laying a foundation for future research.

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Keywords: Microbial eukaryotes, protists, single-cell sequencing, long-reads metabarcoding, peatlands

P-76: Anti-leukemic action of new protein kinase CK2 inhibitors

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Increased expression of protein kinase CK2 is a hallmark of many hematological malignancies, directly promoting proliferation and allowing leukemia cells to evade apoptosis, making it a crucial therapeutic target. This study aimed to investigate the molecular mechanisms of two novel CK2 inhibitors: 1-phenyl-3-(4,5,6,7-tetrabromo-2-methyl-1*H*-benzimidazol-1-yl)propan-1-one (**1a**) and 1-[(4,5,6,7-tetrabromo-1*H*-benzimidazol-2-yl)sulfanyl]propan-2-one (**2a**) in acute lymphoblastic leukemia (CCRF-CEM) and chronic myeloid leukemia (K-562) cell lines.

The research evaluated the effectiveness of these compounds and their reference compounds: 4,5,6,7-tetrabromo-2-methyl-1*H*-benzimidazole (2Me-TBBI, **1**) and 4,5,6,7-tetrabromo-2-(methylsulfanyl)-1*H*-benzimidazole (K37, **2**), in reducing cell viability. A key focus was assessing the ability of the new derivatives to inhibit intracellular CK2 kinase activity, monitored by analyzing the phosphorylation status of specific protein markers using phosflow cytometry and western blot methods. Detailed analyses of the molecular mechanisms of cell death were conducted, including the assessment of cell cycle progression and identification of apoptotic markers.

The results demonstrated that the newly synthesized derivatives, especially compound **1a**, effectively reduce the viability of leukemic cells and exhibit strong inhibitory activity against intracellular CK2 kinase. The distinct molecular response of the cell lines was evident through effective intracellular inhibition and pronounced induction of apoptosis in the CCRF-CEM line, and effective inhibition accompanied by cell cycle arrest and initiation of apoptotic events in the K-562 line.

The ability of these compounds to inhibit CK2 and modulate survival pathways highlights their strong therapeutic potential as promising candidates for the development of anticancer therapies.

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Keywords: leukemias; protein kinases, CK2 kinase, CK2 kinase inhibitors, 4,5,6,7-tetrabromo-1*H*-benzimidazole

P-77: Identification of *Ochrobactrum anthropi* ATCC49188 genes essential for the colonization of biotic and abiotic surfaces

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Ochrobactrum anthropi (Brucellaceae family) is a cosmopolitan species widely distributed across various environments, commonly isolated from soil, water or plant rhizosphere. In clinical settings, the bacterium is recognized as an opportunistic pathogen responsible for nosocomial infections (e.g., catheter-associated). *O. anthropi* constitutively produces AHL-degrading enzymes, allowing it to disrupt the QS systems of phytopathogens such as *Pectobacterium* or *Dickeya*. Furthermore, its ability to form biofilms facilitates the colonization of host plants. In this study, a forward genetics approach was employed to generate transposon mutants of *O. anthropi*. The obtained library of mutants was evaluated for their biofilm formation capacity on artificial substrate (polystyrene) and adhesion efficiency to tomato roots. Several mutants have been identified, exhibiting a reduction in biofilm formation of up to 90% compared to the wild-type strain. To quantify adhesion to biotic surface, attachment assays were conducted using tomato seedlings. Colonization intensity of seedlings inoculated with the mutants were analyzed by isolating bacteria from tomato roots. Understanding the mechanisms of biofilm formation is of critical importance; from a medical perspective, it may facilitate the future development of novel strategies to eradicate these antibiotic-resistant bacteria from hospital environments, while from an agricultural perspective, it offers the potential to enhance crop yields by exploiting QQ mechanisms against phytopathogenic bacteria.

Keywords: Ochrobactrum, biofilm, transposon mutagenesis, adhesion

P-78: *Aegeritella superficialis* symbiosis – antagonistic or mutualistic? New light on the ant-infecting fungus 50 years after species description

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Aegeritella superficialis is known as an ectoparasitic fungus, found mainly on ants from the *Formica rufa* group. First reported in 1974 by Bałazy and Wiśniewski from mounds located in Wielkopolski National Park, it was described as the type species for the genus. Based on morphological traits four other species were classified as *Aegeritella*. Later phylogenetic studies revealed the group to be polyphyletic, with species belonging to different phyla. This taxonomic knowledge raises new questions concerning the biology and ecology of this fungus, as still little is known about the life cycle and transmission mechanisms of *Aegeritella superficialis*.

Genetic studies classify *A. superficialis* in the order Capnodiales, which is known to consist of epiphytic fungi associated with honeydew and sap-sucking insects. *Formica* ants form mutualistic interaction with sap-sucking aphids, tending them and harvesting honeydew. This points to a possible role honeydew and aphids may play in the ant-fungal symbiosis.

The aim of this study was to explore the effects of an *Aegeritella superficialis* infection on *Formica polyctena* colonies and determine whether mound size and its surroundings impacted its prevalence.

For this purpose we collected data from 65 *Formica polyctena* colonies, including the mound size, its immediate surroundings and the percentage of infected ants. Furthermore we analysed the number and diversity of myrmecophile invertebrates from each colony.

As a result we found a doubled infection percentage (c.a. 23%) in relation to what Bałazy reported in 1974. Infection levels varied significantly between mounds (SD 23%), suggesting a strong influence of their surroundings. The size and therefore age of the mound did not correlate with the prevalence. These results indicate that local environmental factors may shape infection dynamics.

Further studies on seasonal variation are needed to fully understand this ecological interaction.

Keywords: *Aegeritella*, ecological interactions, ectoparasites, redwood ants

P-79: *In vitro* model of early symptoms of diabetic nephropathy: hyperglycemia induced rapid changes to mitochondrial respiration and biogenesis in renal tubular cells

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The early stages of diabetes are characterized by elevated insulin and glucose levels, which lead to impaired podocyte function and glomerular filtration barrier. Tubular epithelial cells, present in the lining of the nephrons, are also involved and contribute to tubulointerstitial lesions.

Here we aimed to evaluate the very early symptoms of tubular epithelial cells pathology under high glucose conditions to discover new diagnostic markers for early detection of diabetic nephropathy. Epithelial cells have a high ATP requirement. Mitochondria are also considered centers for extracellular signal propagation. Therefore, aerobic respiration, glycolysis and mitochondrial biogenesis were studied in HK-2 cells at various glucose concentrations. The metabolic profiles were assessed using the Seahorse XF Pro Complete System. The time course of high glucose effect on glycolytic enzymes, electron transport complexes (OxPhos) and mitochondrial biogenesis was examined using western blotting. Preliminary metabolic profiling revealed that under normoglycemic conditions (5 mM glucose), HK-2 obtain the majority of its ATP through glycolysis (70%), while 30% is produced in mitochondria. Exposing cells to higher glucose concentrations: 11 mM (SG) and 30 mM (HG) increased the rate of glycolysis after 5 days of treatment. This was accompanied by a decrease in basal and maximal respiratory rates, although protein levels of OxPhos subunits were elevated. Thus, HK-2 cells exposed to higher glucose concentrations exhibit increased glycolytic flux, but signs of mitochondrial damage are observed only in cells exposed to HG.

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Keywords: Diabetes, epithelial cells, mitochondrial respiration, glycolysis, oxidative stress

P-80: N-Methylated Insulin-Derived Peptides as Inhibitors of β -Amyloid Aggregation: A Potential Strategy for Targeting Neurodegenerative Diseases

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Neurodegenerative diseases such as Alzheimer's disease, Parkinson's disease, and amyotrophic lateral sclerosis (ALS) represent a major and growing global health challenge, particularly in aging populations [1]. These disorders are associated with pathological protein aggregation and amyloid deposition in the central nervous system [1]. A key molecular feature is the formation of insoluble β -sheet-rich structures, arising independently of the primary amino acid sequence [2].

This project investigates the potential of N-methylated peptide analogues, derived from amyloidogenic "hot spots" of insulin, as inhibitors of protein aggregation. Due to the universal mechanism of amyloid formation, such inhibitors may act on multiple aggregation-prone proteins, including β -amyloid, α -synuclein, and SOD1 [3,4]. Five N-methylated analogues of the insulin fragment VEALYL were obtained using triazine based condensing reagents: 12VEA(N-Me)LYL17, 12VEAL(N-Me)YL17, 12VEA(N-Me)LY(N-Me)L17, 12VEALY(N-Me)L17, and 12VEA(N-Me)L(N-Me)YL17. Their purity and structure were confirmed by LC-MS. Anti-aggregative activity was evaluated against β -amyloid (1-42) using Congo Red, Thioflavin T assays, and microscopy [5,6].

Further studies include activity toward α -synuclein and SOD1, as well as proteolytic stability, cytotoxicity, and blood-brain barrier permeability [4]. The results may support development of peptide-based therapeutics targeting pathological protein aggregation.

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Keywords: neurodegenerative diseases, protein aggregation, N-methylated peptides, β -amyloid, peptide inhibitors

P-81: Transcriptome-wide mapping of m6A sites in MDA-MB-231 cells

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N6-methyladenosine (m6A) is the most abundant internal RNA modification that plays a key role in regulating RNA metabolism, such as splicing, stability, nuclear export, interactions with other molecules and translation. Its deposition is catalyzed by the methyltransferase complex and METTL3 is its primary and catalytic subunit. Dysregulation of METTL3 changes gene expression programs involved in cell proliferation, apoptosis, and differentiation, demonstrating a context-dependent role in tumorigenesis.

In our project, we investigate the role of m6A in the triple-negative breast cancer cell line MDA-MB-231 by performing the knock-down of METTL3. The knock-down was achieved using METTL3-targeting shRNA delivered via a lentiviral system and validated using qPCR as well as western blot. To map m6A marks, we used direct RNA sequencing that enables identification of RNA modifications in native transcripts without cDNA conversion. We identified the m6A sites at a single-nucleotide resolution. Next, we aim to investigate the impact of METTL3 on gene expression in MDA-MB-231 cells.

Keywords: N6-methyladenosine, epitranscriptomics, breast cancer, direct RNA sequencing

P-82: Phytohormones as a Novel Therapeutic Concept in the Treatment of Triple Negative Breast Cancer (TNBC)

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Triple negative breast cancer (TNBC) is an aggressive and difficult to treat tumor. The lack of estrogen receptor (ER), progesterone receptor (PR), and human epidermal growth factor receptor 2 (HER2) expression makes classical targeted therapies highly ineffective. This leads to low treatment efficacy, short remission, and high mortality. Despite advancements in immunotherapy and chemotherapy, TNBC remains a major unmet clinical need.

This study investigated the therapeutic potential of selected phytohormones, organic compounds regulating plant development, as bioactive agents for TNBC therapy. These hormones are increasingly recognized for modulating oxidative stress, inflammation, and apoptosis in mammalian cancer cells.

The research utilized the human MDA-MB-231 cell line, a well-established TNBC model with high migratory capacity and drug resistance. During the MTT assay cells were exposed for 72 hours to five phytohormones: methyl jasmonate (MeJA), jasmonoyl-isoleucine (JA-Ile), ethephon (ETH), epibrassinolide (EBL), and indole-3-acetic acid (IAA). The MTT assay evaluated cytotoxicity, while confocal fluorescence microscopy visualized morphological changes and cellular organelle structures.

Results showed that the applied phytohormones significantly disrupted TNBC cell metabolic activity, causing a marked, concentration dependent decrease in viability. Microscopic analysis revealed substantial morphological alterations, notably mitochondrial shape disruption and nuclear fragmentation. These changes strongly suggest the induction of pathways inhibiting proliferation and initiating apoptosis. These observations confirm phytohormones profoundly impact TNBC cell physiology, highlighting them as promising natural compounds with anticancer properties. They may become vital components of innovative adjuvant therapies within "green oncology."

Keywords: breast cancer, TNBC, phytohormones, MeJA, IAA, EBL

P-83: Potential of Environmental Yeast Strains in Winemaking

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The selection of local yeast strains is a growing trend in the global wine industry, aimed at enhancing regional sensory profiles and process efficiency, offering brand-new products. This study aimed to isolate and characterize environmental yeast strains from Polish vineyards located in the Masovian Voivodeship, in order to assess their suitability for industrial winemaking, specifically focusing on ethanol tolerance, fermentation kinetics, and flocculation capacity. A total of 32 isolates were obtained from *Vitis vinifera* cv. Regent using standard Sabouraud medium and a modified Sabouraud+ medium. The latter significantly improved isolation efficiency. Strains were characterized by morphological analysis, ITS-RFLP profiling with *HaeIII*, PCR detection of the *FLO1* gene, and flocculation assessment using the standardized Helm test. Identified genera included *Saccharomyces*, *Rhodotorula*, and *Pichia*. *HaeIII* digestion did not enable full intraspecific differentiation. Four isolates exhibited measurable flocculation (up to 77.7%) despite lacking the *FLO1* gene. Three selected strains were subjected to micro-vinification to evaluate fermentation performance based on CO₂-related mass loss. Active fermentation commenced within 48–72 h, with a decline observed after approximately 14 days. Estimated ethanol yields ranged from 7% to 10% ABV. These findings highlight the substantial microbial diversity of Polish vineyards and identify strains with favorable technological traits, including efficient fermentation kinetics and moderate flocculation. The isolates represent promising candidates for the development of region-specific starter cultures to enhance product quality and process reproducibility.

Keywords: wine, yeast, fermentation, flocculation

P-84: Label-Free Electrochemical Monitoring of Microcystin-LR Release from *Microcystis aeruginosa* Using an Electrified Liquid–Liquid Interface

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Harmful algal blooms (HAB) represent a significant global threat to water quality and public health due to the release of potent cyanotoxins. *Microcystis aeruginosa* is one such prominent cyanobacterial species responsible for HAB and the release of one of the most potent toxins, microcystin-LR (MC-LR), highlighting the need for rapid, cost-effective, and reliable monitoring strategies.

In this study, a label-free electroanalytical platform based on the electrified liquid–liquid interface (eLLI) was developed for the direct detection and periodic monitoring of MC-LR released from *M. aeruginosa* cultures over a 35-day growth period. Unlike conventional electrochemical sensors that rely on surface modification or biorecognition elements, this method enables direct MC-LR detection through voltammetric signals generated from interfacial charge-transfer processes, involving adsorption and ion transfer at the interface. The developed eLLI sensor exhibited an analytical sensitivity of $14.28 \text{ A}\cdot\text{M}^{-1}$, with detection and quantification limits of 55 nM and 500 nM, respectively. Electroanalytical results showed a strong correlation with chromatographic measurements, confirming the analytical reliability of the platform in complex biological matrices. This work establishes eLLI as a promising, rapid, cost-effective, and alternative solution for monitoring cyanotoxins. The approach provides a foundation for the development of soft electrochemical sensing systems for real-time environmental water quality assessment and early detection of algal blooms.

Keywords: Electrified liquid–liquid interface (eLLI), Microcystin-LR, *Microcystis aeruginosa*, Electrochemical sensor, Cyanotoxins

P-85: Impact of Purification Method on the Anti-Inflammatory Activity and Green Chemistry of Aspirin Synthesis

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Understanding the impacts of different purification methods of aspirin, a key anti-inflammatory drug, is important when creating the most effective and safe nonprescription medication as well as preventing any potential chemical waste. Aspirin purification techniques affect anti-inflammatory levels and impurities, therefore this study aimed to determine which recrystallisation conditions: acidic (HCl) and ethanol-water mixtures (50:50 and 70:30), was the most biologically effective and environmentally friendly. Aspirin was synthesized through salicylic acid esterification by acetic anhydride and purified via the three mentioned techniques. The greenness of each technique was evaluated by titrating wash filtrates with NaOH to identify acidic impurities and measuring the production yield of aspirin. Efficiency was qualitatively analyzed by a FeCl₃ test to identify unreacted salicylic acid. Anti-inflammatory properties were assessed by an *in vitro* albumin denaturation assay, measured by spectroscopy. The 70:30 ethanol purification demonstrated the lowest acidic impurities in effluent, with titrant values decreasing rapidly after the initial wash. The product tested with FeCl₃ produced the lightest color, highlighting aspirin purity. However, this method exhibited the lowest yield (43.8%) and anti-inflammatory activity (20.1% inhibition) compared to the acidic purification that had both the highest yield (52.8%) and anti-inflammatory activity (72.4% inhibition) despite exhibiting the highest impurity levels. The findings suggest that increased purification did not correlate with enhanced biological effectiveness, potentially due to anti-inflammatory effects of residual salicylic acid higher in acidic and 50:50 ethanol purifications. Also, the 70:30 water-ethanol mixture provides a more effective polarity for selective crystallization. These results highlight a need to optimize purification methods to maximize chemical purity, yield, bioactivity and sustainability.

Keywords: acetylsalicylic acid, recrystallisation, anti-inflammatory activity, green chemistry, protein denaturation assay

P-86: Enhancing Serological Immunoassay Performance via Nanozyme-Functionalized and Microfluidic Systems

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Serological testing plays a central role in diagnosing autoimmune diseases and infections caused by bacterial, viral, and parasitic agents. By enabling the detection of pathogen-specific antibodies in serum or plasma, these assays offer safer and faster alternatives to conventional microbiological methods. Despite the prevalence of serological assays—particularly in ELISA formats—their performance is often limited by analytical challenges, including nonspecific adsorption, high endogenous immunoglobulin levels, and intrinsic peroxidase-like activity. These factors reduce sensitivity, specificity, and reproducibility, and hinder integration into automated and miniaturized systems.

To address these limitations, we propose a novel assay format based on multifunctional magnetocatalytic nanosorbents. The developed system integrates magnetic separation of immune complexes with catalytic signal generation, enabling dual-specific recognition of serological targets, suppression of endogenous interference, and improved analytical performance. Functionalized nanoparticles, conjugated with antibodies or antigens as immunoreceptors, allow selective isolation of target analytes from complex matrices under an external magnetic field. Additionally, their intrinsic catalytic activity provides a stable and cost-effective alternative to conventional enzyme labels, reducing assay complexity and enhancing stability across varying environmental conditions.

The proposed approach enables simplified assay protocols and supports integration into compact, user-friendly diagnostic platforms. By combining selective purification with robust colorimetric detection, it offers a promising route toward sensitive, reliable, and point-of-care diagnostic systems.

Keywords: magnetic nanoparticles, nanozymes, ELISA, serology, analyte separation

P-87: Photomodulated effect of graphene oxide and chlorophyll-based substrates on the morphological and functional status of U87 glioma cells *in vitro*

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Glioblastoma multiforme is the most common and the most aggressive brain tumor. The average incidence of this tumor ranges from 3.19 to 4.17 cases per 100,000 people-year (around 48% of all CNS malignancies), while median survival time is only 15 months. Only 3-5% of patients survive 3 years after diagnosis. Currently known and used therapy for glioma is characterized by limited effectiveness, serious side effects, and in most cases, only offer limited extension of patient survival. Furthermore, progress in treatment remains insignificant, with little improvement in survival rates over the last 30 years. Due to this, the search of new treatment methods remains relevant. One of possible solutions is targeting oxidation phosphorylation (OXPHOS) to alter cancer metabolism. We hypothesize, that this can be achieved by delivering light energy to cell through oxide-chlorophyll layer.

The aim of this study was to investigate the effect of graphene oxide, chlorophyll, and graphene oxide-chlorophyll fusion on the morphological and functional status of U87 glioma cells *in vitro*. Cells were incubated in plate wells coated with the appropriate reagent and exposed (vs. not exposed) to LED radiation. Changes were assessed using the PrestoBlue viability assay and morphology, observed using a light microscope and SEM. Significant reductions in viability and morphological abnormalities were observed in the irradiated group cultured on a chlorophyll layer and in the irradiated control group. An increase in viability was observed in the irradiated group cultured on a graphene oxide-chlorophyll composite layer. These findings suggest that the use of chlorophyll, chlorophyll with light irradiation and irradiation itself may have potential in therapeutic use against glioma.

Keywords: U87 cell line, graphene oxide, chlorophyll, photon energy transfer, nanoparticles.

P-88: From Surface to Sensor: Optimizing Screen-Printed Electrodes for Biosensing Applications

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The development of low-cost, miniaturized electrochemical biosensors for point-of-care (POC) applications relies heavily on the properties of the transducer surface used to create receptor layer. Screen printed (SP) electrodes offer a particularly attractive solution due to their scalability, low fabrication cost, and ease of integration into microfluidic systems. However, their surface composition often contains residual polymeric binders and can significantly influence the formation and stability of self-assembled monolayers (SAMs), which are essential for effective biomolecular recognition.

In this study, we investigated the effect of various surface cleaning protocols on the physicochemical properties of SP gold electrodes and their suitability for SAM formation. Surface characterization revealed that more aggressive cleaning procedures effectively reduced the presence of polymer residues, exposing a greater fraction of underlying metal grains. This modification resulted in a more favorable surface morphology for thiol-based SAM assembly. Electrochemical and functionalization studies further demonstrated that these treated surfaces enabled more efficient receptor immobilization, leading to improved surface coverage and biosensor performance.

Additionally, we evaluated the long-term stability of the functionalized biosensor surfaces under storage in mercaptohexanol (MCH) solutions of varying concentrations. The results indicate that appropriate selection of MCH conditions allows preservation of the receptor layer integrity and functionality for up to three months, highlighting a viable strategy for extending sensor shelf life.

Overall, these findings demonstrate that optimized surface treatment and storage conditions can significantly enhance the performance and practicality of SP electrode-based biosensors. This supports their potential as robust, cost-effective platforms for integration into portable microfluidic POC devices.

Keywords: Electrochemical biosensor, printed electronics, surface functionalization, screen printing technology

P-89: Tracking the drop: Investigating erythroid ACKR1 in the Duffy-null neutrophil mystery

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The Duffy-null blood type, highly prevalent in individuals of African descent, results from the genetic variant rs2814778(G), which disrupts the GATA1 motif in the ACKR1 promoter and prevents Duffy-antigen (aka ACKR1) expression on red blood cells (RBCs). In addition, Duffy-null individuals often exhibit a lower neutrophil count, a condition known as Duffy null-associated neutrophil count (DANC). DANC is frequently misclassified as neutropenic, leading to unnecessary medical interventions, altered treatment plans, and exclusion from clinical trials.

To investigate the impact of this ACKR1 polymorphism on health and disease, we generated *Ackr1*(GATA1-G) mice, mimicking the human rs2814778(G) variant. These mice lack ACKR1 on RBCs but retain endothelial expression. Moreover, *Ackr1*(GATA1-G) mice exhibit reduced blood neutrophil counts, establishing them as a model for studying DANC.

We then assessed whether DANC results from impaired neutrophil production or mobilization. Bone marrow granulopoiesis was unchanged in *Ackr1*(GATA1-G) mice compared to wild-type control mice. Moreover G-CSF treatment did not show an alteration in the number of circulating neutrophil in *Ackr1*(GATA1-G) mice. Together these data indicate that neutrophil production and release remain intact in the absence of ACKR1 on erythroid cells. However, *Ackr1*(GATA1-G) mice displayed an increased neutrophil recruitment to the peritoneal cavity following CXCL8 injection, suggesting that reduced blood neutrophil counts results from an altered trafficking and an enhanced tissue infiltration.

In summary, this new model provides valuable tool for studying ACKR1-mediated neutrophil homeostasis and may refine clinical interpretations of neutrophil counts in Duffy-null individuals.

Keywords: chemokines, Duffy-null, neutrophils,

P-90: Graphene Quantum Dots Modulate Photosynthetic Pigments Without Affecting Growth in *Lepidium sativum*

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Graphene quantum dots (GQDs) are novel nanomaterials, yet their effects on plants are not well understood, particularly regarding their toxicity and physiological effects. This study evaluated the impact of GQDs on the germination and early growth of *Lepidium sativum*, a model plant, at concentrations ranging from 0 to 500 ppm.

GQDs were synthesized using a green citric acid method. Seeds were placed in six-well plates and exposed to GQD solutions in distilled water, along with an untreated control. After 7 days of incubation, root length, shoot length, fresh weight, and photosynthetic pigment content (chlorophyll a, chlorophyll b, and carotenoids) were measured.

The synthesized GQDs displayed fluorescence at 380 nm excitation and 520 nm emission, with an average size of about 5 nm. Root length increased at select concentrations compared to the control, while shoot length, root-to-shoot ratio, and fresh weight did not change significantly. Chlorophyll a content increased, while chlorophyll b remained constant, and elevated carotenoid levels altered the ratio. Despite the increase in carotenoids, indicating potential stress, no negative impact on plant growth was observed, suggesting a mild adaptive response.

In summary, GQDs at the tested concentrations were not toxic to *Lepidium sativum* and may enhance specific physiological parameters. These findings increase the understanding of plant-nanomaterial interactions and encourage further research.

Keywords: GQD, *Lepidium sativum*, nanomaterial, toxicity

P-91: *Pontechium maculatum* (L.) Böhle&Hilger as a model for *in vitro* culture establishment and conservation of a steppe relict species

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Pontechium maculatum (L.) Böhle&Hilger is a rare, biennial species belonging to the Boraginaceae family, naturally occurring in dry grasslands from Central Europe to Western Asia. It is considered a steppe relict with a pontic distribution, reflecting its persistence from warmer and drier climatic periods of the Holocene. Due to its limited occurrence and habitat fragmentation, this species is regarded as endangered, which highlights the need for effective conservation strategies.

The phytochemical profile of *P. maculatum* is characteristic of the Boraginaceae family and includes biologically active compounds such as phenolic acids, phytosterols, and naphthoquinones, including shikonin derivatives. These compounds exhibit antioxidant, anti-inflammatory and antimicrobial properties, as well as the ability to stimulate tissue regeneration. At the same time, the presence of potentially toxic metabolites typical for this family necessitates controlled approaches to their study and utilization.

The aim of the study was to develop an *in vitro* culture system for *Pontechium maculatum* as a tool for both ex situ conservation and future biotechnological applications. Particular attention was given to the initiation of cultures under aseptic conditions and the evaluation of explant response in terms of regeneration potential. The establishment of stable *in vitro* cultures may provide an alternative source of valuable secondary metabolites and contribute to the protection of this rare species.

The obtained results indicate that *P. maculatum* exhibits promising regenerative capacity under *in vitro* conditions, supporting its potential as a model species for further studies on controlled biosynthesis of bioactive compounds.

Keywords: *Pontechium maculatum* (L.) Böhle&Hilger, *in vitro* cultures, plant conservation, Boraginaceae, secondary metabolites

P-92: Bioinformatic analysis and optimization of recombinant HuFABP7 protein production in *Pichia pastoris*

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Fatty acid-binding proteins (FABPs) are a group of proteins especially active in tissues with high fatty acid metabolism. To this family belongs FABP7 (B-FABP) – a protein expressed in the brain mostly during brain development. This protein is crucial for neuronal differentiation thanks to its binding and trafficking of polyunsaturated fatty acids, and its expression is very low in adults. Research shows that in some brain diseases, like Alzheimer's disease, amyotrophic lateral sclerosis, and malignant glioma, there is a correlation between higher levels of FABP7 and poor prognosis for patients' recovery. The aim of this study was to perform bioinformatic analysis and validate the production of recombinant FABP7 protein in the yeast expression system *Pichia pastoris*. For this research, the pPICZ α A plasmid with protein sequence was optimized for replication in *Escherichia coli* and then protein production in *Pichia pastoris*. The optimized procedure of obtaining the FABP7 protein is a primary step to later examine and understand the inflammation and how B-FABP contributes to the activation of cells of the immune system. That may lead to the answer if FABP7 can be a marker for early recognition of some serious neurological disorders.

Keywords: FABP7; human recombinant protein; yeast expression system; bioinformatic analysis

P-93: The effect of CHK1 kinase inhibitor and aldehyde dehydrogenase inhibitor on OVCAR8 ovarian cancer cells

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Ovarian cancer is considered one of the most important global challenges in medicine. According to GLOBOCAN 2022 data, ovarian cancer is the eighth most common cancer and the eighth leading cause of death among women worldwide. Metastases to the abdominal peritoneum, among other places, significantly hinder surgical reduction of tumor mass. Chemotherapy for ovarian cancer is usually based on platinum compounds and taxanes. Currently used therapeutic methods do not achieve the intended goal of completely eradicating the cancer.

The aim of this study is to investigate the action of drugs targeting the repair mechanism of cancer stem cells. Checkpoint 1 kinase inhibitors (CHK1i) together with disulfiram comprehensively target the cell repair system. Disulfiram, an example of an ALDH1 inhibitor known from alcoholism therapy, causes DNA damage resulting from the accumulation of aldehyde. In turn, the CHK1 inhibitor (CHKi, MK8776) induces DNA damage by blocking the repair of genetic material that occurs during cell cycle arrest.

The cytotoxicity of disulfiram and MK8776 (CHK1i) was determined in studies using both monotherapy and combination therapy to determine the coefficient of drug interaction (CDI) using the MTT cytotoxicity test. The ability of cells to form colonies was also estimated using the clonal growth method. The studies were conducted on the OVCAR8 line, which exhibits epithelial morphology. After applying appropriate concentrations of the tested drugs, a decrease in the viability of OVCAR8 cells was observed in the MTT cytotoxicity test, as well as a decrease in the ability of cells to form colonies. Additionally, the results from both methods indicate that combination therapy is more effective than monotherapy with disulfiram or MK8776. Targeting population of stem cells and using drug combinations may have beneficial effects and provide valuable information needed to combat the very dangerous adversary that is ovarian cancer and cancer in general.

Keywords: CHK1 kinase inhibitor, disulfiram, aldehyde dehydrogenase, ovarian cancer, stem cells

P-94: Structural and functional characterization of *pco* and *sil* copper resistance regions in *Cronobacter sakazakii*

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Copper participates in the immune system's response to infection – its accumulation within macrophages leads to the inactivation of pathogens. Consequently, copper resistance in bacteria is a significant virulence factor. The most common resistance mechanism is the efflux of the toxic element outside the cell. These efflux systems include the *pco* and *sil* resistance modules.

In this study, the *pco* and *sil* regions in bacteria of the genus *Cronobacter* were characterized and found to be located chromosomally (*Cronobacter sakazakii* LMG 2762) or on a mobile genetic element (the pCS-MK10_P2 plasmid of *C. sakazakii* MK10). In strain LMG 2762, *pco* and *sil* are located within the putative Tn7 transposon. LMG 2762 and MK10 strains contain both of these copper resistance modules.

In this research, bioinformatic analyses were performed, including the identification and genetic organization of the *pco* and *sil* modules, as well as that of Tn7. A comparative analysis of chromosomally and plasmid-located modules in both *Cronobacter* strains was also conducted.

Furthermore, the widespread distribution of copper resistance genes in the genomes of other *Cronobacter* strains and bacteria within the family *Enterobacteriaceae* was demonstrated. Preliminary functional analyses of the selected *pco* and *sil* modules showed no significant differences in minimum inhibitory concentration (MIC) values between copper-resistant and non-resistant strains under aerobic conditions. This suggests that further research should also examine MIC values under microaerophilic and anaerobic conditions.

The localization of copper resistance genes within mobile genetic elements facilitates their dissemination, especially under selective pressure. Many plant protection products are based on copper compounds. *C. sakazakii* is an environmental bacterium and a human pathogen. The acquisition of resistance genes, important for pathogenesis, may have occurred through horizontal gene transfer in the environment.

Keywords: *Cronobacter sakazakii*; copper; *pco*; *sil*; Tn7

P-95: Impact of NETs on mitochondrial function in ECTV-infected cells – preliminary study

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Neutrophils are the most abundant leukocyte population in human peripheral blood and act as the first line of defense. The protective mechanisms are phagocytosis, extracellular degranulation, and the formation of neutrophil extracellular traps (NETs). NETs are web-like structures composed mainly of chromatin and proteins derived from neutrophil granules. Those structures immobilize and eliminate microorganisms; however, their formation is also associated with the induction of non-infectious inflammatory processes. Although NETs have been extensively studied in antibacterial responses, their role in viral infections remains poorly characterized. The study aimed to evaluate the effects of NETs on mitochondria of the murine L929 fibroblasts infected with ectromelia virus (ECTV) as an *in vitro* model. The source of NETs for further evaluation was neutrophils isolated from human peripheral blood buffy coats. L929 cells were infected with ECTV in the presence or absence of NETs. The mitochondrial network, as well as mitochondrial mass (MMP), mitochondrial membrane potential, and reactive oxygen species level, were assessed in cells using immunofluorescence microscopy and flow cytometry, respectively. Cells exposed to NETs exhibited an altered mitochondrial network morphology, appearing more disorganized and irregular. Flow cytometry results indicate mostly decreased mitochondrial mass and MMP, independent of ECTV infection. These changes were accompanied by increased levels of reactive oxygen species in the presence of NETs. The results suggest a potential role of NETs in modulating mitochondrial function during viral infection.

Keywords: NETs, L929, mitochondria, ECTV

