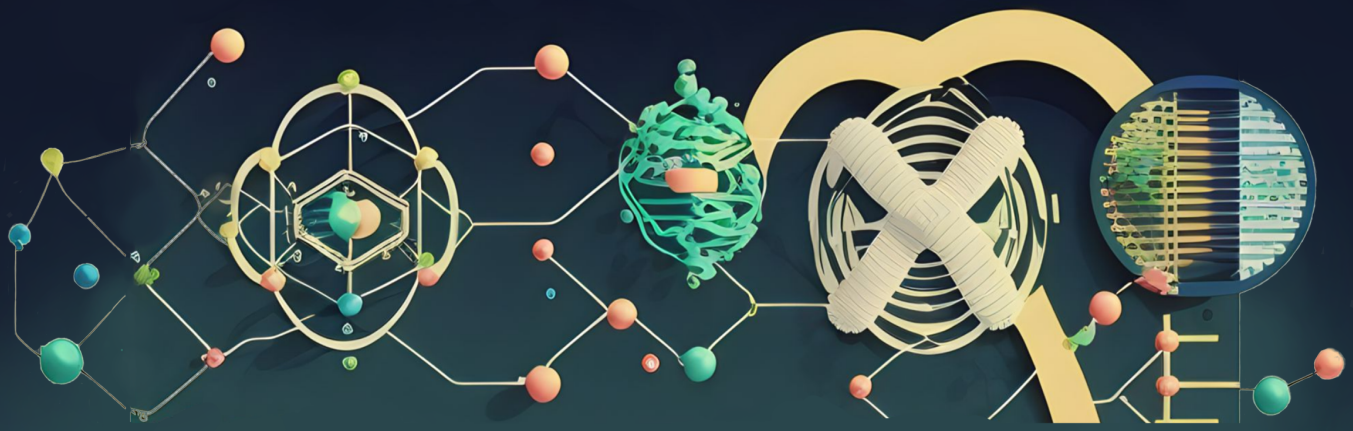
Postdoc and PhD RETREAT

Organised by CEITEC Postdoc and PhD committees



BOOK OF ABSTRACTS

26 –27 October 2023

OREA RESORT DEVET SKAL, CZECHIA

Welcome Address

Dear participants!

We are thrilled to extend a warm and formal welcome to the **CEITEC Postdoc and PhD Retreat**, a collaborative endeavour organized by the PhD students and Postdocs of the Central European Institute of Technology (CEITEC).

In this year's instalment, we are bringing together a diverse assembly of students and post-doctoral scientists hailing from various scientific domains. Anticipate a program filled with enriching highlights, including lectures delivered by accomplished CEITEC researchers, a captivating discourse on "Engineering the Conversation with AI," and an exclusive AI workshop tailored to our postdoctoral colleagues. Our agenda also features two illuminating talks on technology transfer and a session on building bridges, an experience at the University of Edinburgh.

Throughout this two-day event, you can expect to be immersed in a tapestry of nearly 100 contributions from our students and postdocs, ranging from talks to posters. These contributions will span a wide spectrum of academic disciplines, encompassing **life sciences, physics, chemistry, and material sciences.**

This gathering promises a remarkable opportunity to share and delve into the intricacies of cutting-edge research, foster mutual inspiration, and forge valuable connections and meaningful friendships within a safe and convivial environment.

Finally, our deepest gratitude goes out to you for making the CEITEC Postdoc and PhD Retreat a resounding success. Your collective efforts are greatly appreciated.

With warm regards,
Your organising committee,
Pavel Payne
Khadija Hajji
Mateo Seoane Blanco
Oleksii Laguta
Kateřina Linhartová
Lenka Dostálová
Jorge Navarro
Anna Cherian
Kaushik Baishya
Radhika Nittoor Veedu
Jana Juráková
Katarina Novčić

Table of Contents

Program

OCTOBER 26th		
10:00	Registration and welcome coffee	
10:30	Welcome word by PAVEL TOMANČÁK	
11:00	Sponsor talk: Beckman Coulter	
11:15	Invited speaker: LUCY VOJTOVÁ Advanced Nanostructured Biomaterials: From Synthesis, Structure-Property Relationships to Processing and Applications in Regenerative Medicine	
11:50	Lunch	
13:00	Invited lecture: MARTIN SVOBODA Engineering the Conversation with AI	
14:30	Coffee break	
15:00	JAKUB HOLOBRÁDEK Spin waves in synthetic antiferromagnetic	MARTIN SVOBODA AI workshop for postdocs
15:15	MICHAELA PEŠOVÁ Distinct p53 Phosphorylation Patterns in Chronic Lymphocytic Leukemia: Where They Come from and How They Affect p53 Function	
15:30	VAISHALI PANKAI Discovery of novel HDAC inhibitors for breast cancer treatment using computational approaches	
15:45	ANNA CHERIAN An RNA editing-independent function of ADAR1 inhibits PKR activation in mice	
16:00	RAMYA CHITTOORY Tungsten trioxide layers fabricated from nanoparticulate suspension with organic binder via brick-and-mortar approach for enhancing the current density in PEC cell under visible light	
16:15	MICHAELA RŮŽIČKOVÁ Genomics of multi-resistant Escherichia coli circulating in a colony of gulls: dynamics of colonizing strains displays importance of longitudinal wildlife studies	
16:30	INDERJEET BHOGAL Molecular modeling approaches in the identification of novel RAGE inhibitors for the treatment of Alzheimer's disease	DENYS BIRIUKOV Molecular Simulations of Biological Systems: Facts, Myths, and Examples
16:45	SURENDRA SADDALA Unravelling the Molecular Function of CDM1 Zinc-finger Protein in Meiotic Progression in Arabidopsis thaliana	PETR FAJKUS Intricate evolution of Telomerase RNA
17:00	SANAM GAREHBAGHI Spectrophotometric Detection of Calcium and Phosphorous in Human Bone Sample, a Straightforward Method in Clinical Analysis	SAMER HALABI Investigating the Mechanisms of Host-Pathogen Arms Race between rabbits and Myxoma virus
17:15	DENIS ŠUBERT Guanine Quadruplexes as Targets for the Architectural Protein CTCF	MARIE JAKEŠOVÁ Wireless optoelectronic neural stimulators
17:30	Poster session	
19:00	Dinner	
20:30	Pub quiz, karaoke, …	

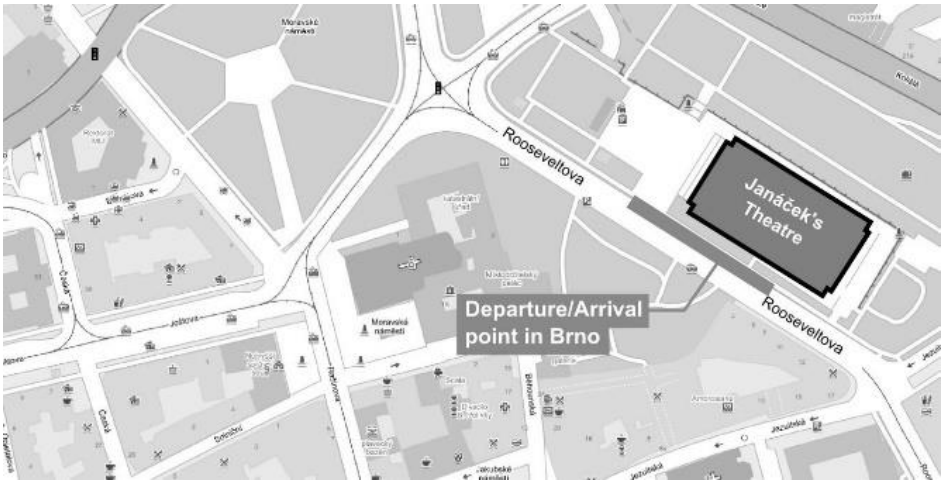
OCTOBER 27th	
8:00	Breakfast
9:15	Jaroslav Koča Bridge Fund: PETER PAJTINKA Development of structured substrate for the study of protein curvature sensing on supported lipid membrane
9:30	Jaroslav Koča Bridge Fund: EVA ČERNÁ Gum Karaya based hydrogels for phage therapy of chronic wounds
9:45	Tech transfer: HAROLD DE VLADAR - Ribbon Biolabs
10:30	Tech transfer: JAN NEUMAN - NenoVision
11:15	Coffee break
11:45	MICHAELA MUSILOVÁ Grant opportunities for early-career researchers
12:00	SAMER HALABI Building Bridges: BioDocSoc's Mission in Edinburgh's School of Biology
12:15	Closing remarks
13:00	Lunch
14:00	Afternoon hike

Information

Transportation

From Brno to Devět Skal →

On Thursday, Oct. 26th, the bus from Brno will be leaving from Rooseveltova street (next to Janáček theatre), at 8:00 am sharp (as indicated in the map below). We kindly ask you to be there at least 10 minutes before the departure. The approximate time of arrival at the venue is 10:00 am.



From Devět Skal to Brno →

The bus from Devět Skal to Brno (arriving at Rooseveltova street) will be leaving the conference venue on Friday, Oct. 27th at 16:00.

Accommodation and meals

Accommodation for the participants will be provided in double rooms in the hotel, which is also the conference venue. We did our best to follow the accommodation preferences.

Breakfast, lunch, and dinner will be served in the Hotel's restaurant. During lunches and dinner, water is provided by the organisers. There is a possibility to order other drinks, but they will not be paid by organizers. Coffee breaks will be served in front of the conference room. Coffee, tea, and snacks will be provided during coffee breaks.

Posters

Please check the number of your poster in the abstract book and match it with the number on the stand. The poster session will take place on Thursday evening before dinner. The participants are encouraged to mount their posters during a lunch break or one of the coffee breaks. The latest before 5:30 pm. Participants with odd numbers should be present next to their poster in the first half of the poster session, and participants with even numbers in the second half of the poster session.

After program on Thursday evening:

After dinner, we prepared the Retreat version of a pub quiz, taking place in the restaurant. The winning team will get a small prize. A limited amount of beer and wine will be available. After the pub quiz, there will be karaoke or space for other activities.

Afternoon hike on Friday:

On Friday, after lunch, everyone is invited to participate in an afternoon hike around the venue. The program may be changed based on the weather.

Welcome talk

Pavel Tomančák

Executive Director of CEITEC

Pavel is a renowned expert in evolutionary and developmental biology and has been a research group leader since 2005 at the Max Planck Institute for Molecular Cell Biology and Genetics in Dresden. His laboratory at the Max Planck Institute focuses on the study of regulation and evolution of gene expression during the embryonic development of multi-cellular organisms. He continues to lead this research group also as a director of the CEITEC consortium. His research is unique among others in that it combines advanced molecular biology with state-of-the-art microscopy techniques and advanced computational analysis of microscopic images. Pavel Tomančák also has a very close relationship with the city of Brno, because it was here that he began his scientific career as a student of molecular biology and genetics at Masaryk University.

Invited speaker

Lucy Vojtová

CEITEC - Central European Institute of Technology, Brno University of Technology, Czech Republic

Advanced Nanostructured Biomaterials: From Synthesis, Structure-Property Relationships to Processing and Applications in Regenerative Medicine

Associate professor L. Vojtová, the leader of the Advanced Biomaterials group at CEITEC Brno University of Technology will present an overview of the group interest and research focused primarily on the design and synthesis of “smart” high-performance stimuli-responsive functional polymers and composites for biomedical applications. She will also highlight hydrogel self-assembly, surface nanopatterning, sol-gel transitions, release and lifetime-controlled biomaterials serving as drug carriers or scaffolds for hard and soft tissue regeneration. Materials processed via rapid prototyping, freeze-drying, or spinning will also be discussed in terms of biological in vitro, ex ovo, ex vivo, or in vivo evaluation. She hopes to provide a flavour of the research her group is performing and to spark your interest in potential future collaborations.

Acknowledgement:
The work is co-financed with the state support of the Technology Agency of the Czech Republic as part of the National Center of Competence II Program No. TN02000017/003, the European Regional Development Fund – the project Mechanical engineering of biological and bio-inspired systems (no. CZ.02.01.01/00/22_008/0004634), and the ProfiBONE project (no. TO01000309) that benefits from a grant from Iceland, Liechtenstein and Norway through the EEA Grants and the Technology Agency of the Czech Republic.

Tech transfer presentations

Jan Neuman

NenoVision, CEO and co-founder

I graduated in Physical Engineering at the FME BUT, which gave me a foundation for a better understanding of physics and instrument design. During my PhD studies, I was actively involved in creating the CEITEC. In 2015, I co-founded NenoVision, the first spin-off at CEITEC. We work on commercializing a special module for electron microscopes that greatly enhances their analysis capabilities. As co-founder and CEO, I strive to push the boundaries of correlative microscopy and succeed commercially with our innovative products.

NenoVision tech transfer story – motivation, experiences and mistakes

NenoVision company is the first CEITEC Spin-off company that commercialized module-extending electron microscopes. Based on our personal experiences, I would like to open the topics and terms fundamental for the entrepreneurs and technology transfer process, such as lean canvas, business plan, product marketing, IP protection, and others. I will also discuss our inner motivation to commercialize products and start the company.

Harold de Vladar

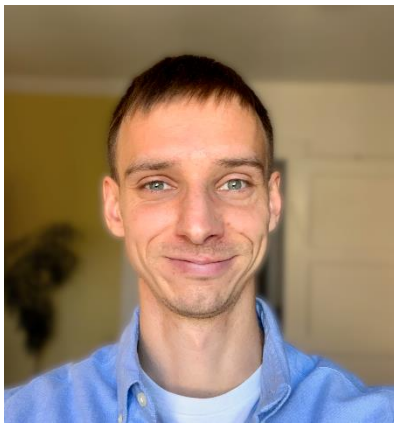
Ribbon Biolabs, CEO and founder

Biotech entrepreneuring: a personal voyage

I want to share my experience and decision-making process that led me to become a biotech entrepreneur by founding, leading and growing Ribbon Biolabs. I was originally a researcher in evolutionary genetics and took the challenge and risk of establishing a new technology for DNA synthesis and build a company around it. The hurdles were – and continue to be – many. How did my science background prepare me for this journey and how did it help me to go forward? It has been an exciting and intense journey, learnings that we never get in a course or project and, above all, a change in the mindset.

AI Lecture and Workshop

Martin Svoboda



Dr. Martin is a physicist and programmer from the University of Jan Evangelista Purkyně in Ústí nad Labem. With a PhD in computer modelling in science and technology, he has spent the last year exploring Artificial Intelligence applications. Martin is part of a group specialising in granular materials simulation and also contributes to the "AI Reactor," a university initiative to promote AI use in an academic environment.

Engineering the Conversation with AI

The presentation begins by demystifying the architecture and capabilities of modern large language models, setting the stage for understanding their application in research and everyday tasks. Attention then shifts to the art and science of 'Prompt Engineering,' where we explore the nuances of eliciting desired responses from AI systems. By showcasing a range of AI tools that utilize these models, attendees will gain insights into how these technologies can be incorporated into academic and research environments. The talk concludes with a discussion on the ethical landscape surrounding language models, including issues of bias and responsible AI usage.

Oral presentations – Postdocs

T1: Molecular Simulations of Biological Systems: Facts, Myths, and Examples

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Molecular dynamics (MD) simulations have progressively emerged as an indispensable scientific tool frequently employed in the field of biology, acting as a bridge between experimental observations and theoretical predictions. However, despite their profound capabilities, the full potential of molecular simulations remains largely untapped. This limitation can be attributed to the dichotomy of perception. Biologists, deeply immersed in the intricate complexities of living organisms, and simulation people, primarily physicists, often view biological systems through different lenses, leading to variances in studied time and length scales. This divergence eventually hinders the integration of simulations into experimental biology. This presentation aims to elaborate on this disconnect by showcasing a series of examples encompassing the modeling of biological systems of various complexity and composition. We will explore (i) how we can model cellular membranes characteristic of various species like mammals, bacteria, and viruses; (ii) which components constitute these membranes, e.g., lipids, proteins, and carbohydrates, and how we can target them for designing novel antibiotics such as antimicrobial peptides; and (iii) how physical concepts ingrained in molecular simulations can foster the rational design of these peptides. All these examples attempt to underscore the true potential of MD simulations across diverse biological disciplines, thereby promoting their wider application and further deepening our understanding of intricate biological phenomena.

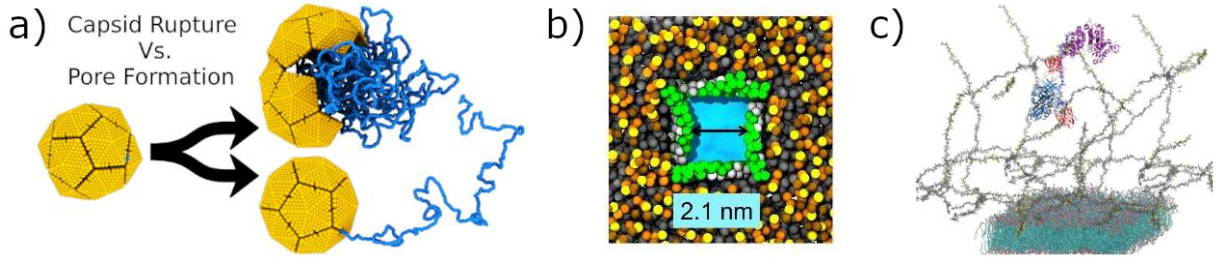

Figure 1. Examples of molecular simulations of biological systems: a) Possible mechanisms of cargo release from virus-like nanoparticles.[1] b) Top-view on a lipid membrane with a barrel pore formed and stabilized by α-helical peptides.[2] c) Proteins captured by polysaccharide chains of a transmembrane syndecan protein.[3]

References

[1] Sukenik, L., Mukhamedova, L., Procházková, M., Škubník, K.; Plevka, P., Vácha, R. *ACS Nano* **2021**, *15*, 19233-19243.
[2] Deb, R., Kabelka, I., Přibyl, J., Vácha, R. *biorxiv* **2023**, https://doi.org/10.1101/2022.05.09.491086.
[3] Biriukov, D., Riopedre-Fernandez, M., Martinez-Seara, H. *manuscript in preparation*

T2: Intricate evolution of Telomerase RNA

P. Fajkus,*[a,c] V. Peška,[a] A. Kilar, ADL. Nelson[b], J.Fajkus*[c]
+ other contributors in reference list

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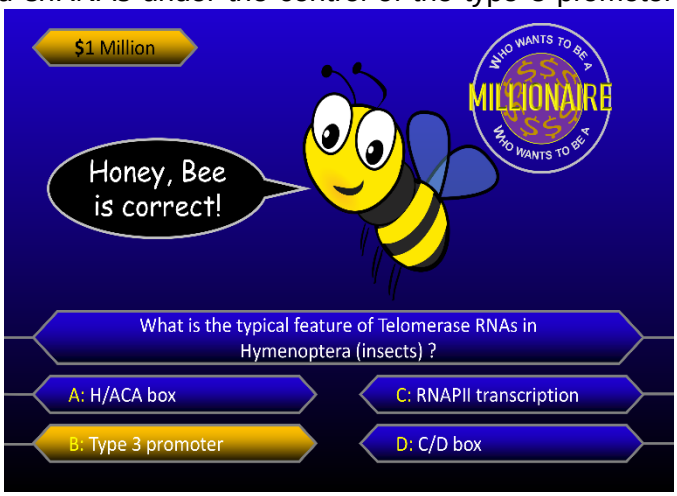
Since the emergence of the first eukaryotes telomere DNA, telomerase and accessory proteins have been key players in ensuring genome stability. Thus, change in any of these players may cause serious consequences for the whole game. On the other hand, game strategies, which had evolved in different eukaryotes are surprisingly diverse - employing variable telomere binding proteins, telomere repeats, telomerase biogenesis pathways, or mechanisms of telomere maintenance without telomerase. Most of this variability stems from the most enigmatic player - telomerase RNA (TR) – which, as a team maker provides, a scaffold for the assembly of the other telomerase components and dictates the sequence synthesized by the telomerase complex.

Characterization of telomerase RNAs represented a challenging task. However, thanks to exhaustive availability of novel genomic data across eukaryotes we developed a computational strategies for TR prediction, which was successfully utilized in characterisation of TR genes across plants **(1)** and early diverged taxa from Diaphoretickes megagroup suggesting a common origin of Ciliate and Plant TRs as PolIII-transcribed snRNAs under the control of the type 3 promoter **(2)**. While Animalia TRs are known as PolII-transcribed H/ACA box snoRNAs, recent identification of previously unknown TRs in insects shown their evolutionary switch to plant-like TRs **(3)**. These results substantially changed previous paradigms in TR evolution and brought a new question about the origin of the TRs.



Graphical abstract

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2. Fajkus, P., Kilar, A., Nelson, A.D.L., Hola, M., Peska, V., Goffova, I., Fojtova, M., Zachova, D., Fulneckova, J. and Fajkus, J. (2021) Evolution of plant telomerase RNAs: farther to the past, deeper to the roots. *Nucleic acids research*, **49**, 7680-7694.
3. Fajkus, P., Adamik, M., Nelson, A.D.L., Kilar, A.M., Franek, M., Bubenik, M., Frydrychova, R.C., Votavova, A., Sykorova, E., Fajkus, J. *et al.* (2023) Telomerase RNA in Hymenoptera (Insecta) switched to plant/ciliate-like biogenesis. *Nucleic acids research*, **51**, 420-433.

T3: Investigating the Mechanisms of Host-Pathogen Arms Race between rabbits and Myxoma virus

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[a]*Institute for Immunology and Infection Research, University of Edinburgh, Edinburgh, United Kingdom*
[b]*Department of Pathology, University of Cambridge, Cambridge, United Kingdom*
[c]*Department of Veterinary Medicine, University of Cambridge, Cambridge, United Kingdom*

Myxoma virus (MYXV) is a poxvirus that infects rabbits and is similar to many other poxviruses that infect humans, such as the chickenpox virus and the monkeypox virus of emerging concern. MYXV was used in Australia in 1950 as a pest control against the European rabbits that were introduced to the country almost two centuries earlier. The use of MYXV then, serves as a large-scale experiment to better understand the host-pathogen arms race. Investigating the genetic variation in historical and modern populations of rabbits showed allele frequency changes of MHC-I genes after the introduction of the MYXV. In our study, we are investigating the genetic and structural changes between the disease-resistant and disease-susceptible MHC-I alleles to understand the impact of the introduction and evolution of the MYXV strains on the evolution of the host MHC-I alleles in rabbits.

Figure 1 Rabbits around a waterhole during myxomatosis trials, Wardang Island, South Australia, 1938.

T4: Wireless optoelectronic neural stimulators

M. Jakešová,[a] M.J. Donahue,[b] M. Silverå Ejneby,[b] A. Caravaca,[c] P. Olofsson,[c] and E. D. Głowacki*[a]

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Implantable electronics used in bioelectronic medicine are normally powered via an implantable battery, which increases the device footprint and further burdens the patient with regular battery replacement surgeries. This motivates the implementation of wireless power transfer solutions which can lead to more minimalistic implants. One of the least-explored routes is photonic power transfer followed by light-to-electricity transduction via an implanted photovoltaic (PV) cell. We show how useful amounts of light energy can be safely transferred through tissue when using wavelengths in the deep red region of the spectrum, 630-700 nm. PVs are power sources which will deliver a given current based on the load impedance. We demonstrate this principle by making PV neurostimulator circuits. We show how various variables like PV arrangement, pulse frequency, and stimulation electrode material affect the stimulation current which can be delivered by a PV neurostimulator circuit. The device concept was validated as a peripheral nerve stimulator on a vagus nerve in a mouse model.[1] The result of our efforts is a minimalistic microstimulation device, which can be altered according to the desired stimulation target and could be highly enabling for *in vivo* small animal experimental protocols.

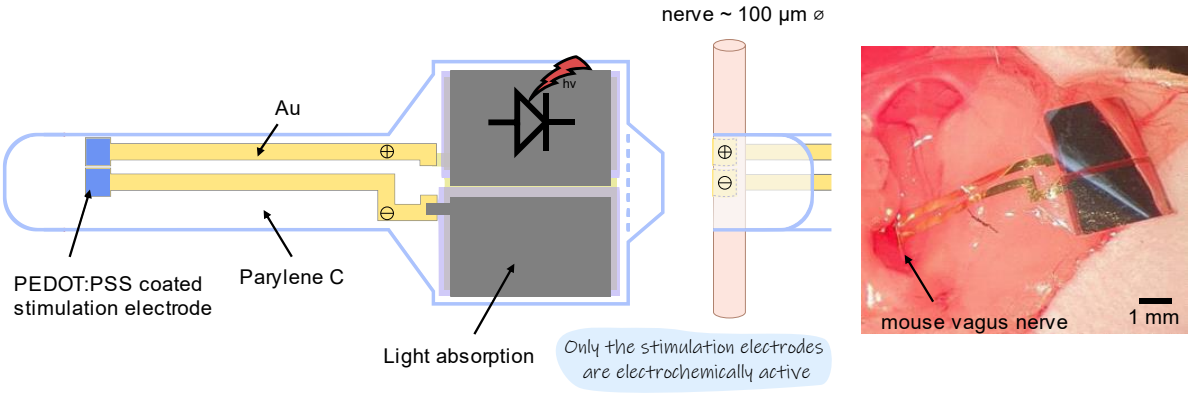

Figure 1. Schematic of a photovoltaic stimulator, a photograph of the device wrapped around a mouse vagus nerve

References

[1] Donahue, M. J., Ejneby, M. S., Jakešová, M., Caravaca, A. S., Andersson, G., Sahalianov, I., Derek, V., Hult, H., Olofsson, P. S., Glowacki, E. D. *J. Neural Eng.* **2022**, 19, 066031.

T5: Building Bridges: BioDocSoc's Mission in Edinburgh's School of Biology

S. Halabi,[a,b]

[a]*Institute for Immunology and Infection Research, University of Edinburgh, Edinburgh, United Kingdom*
[b]*Department of Pathology, University of Cambridge, Cambridge, United Kingdom*

BioDocSoc, or the "Biology Doctoral Society" at the University of Edinburgh, United Kingdom, is a vital organization dedicated to supporting postdoctoral researchers and PhD students in the School of Biology. Its core mission is to foster a sense of community among postdocs and actively engage them in school initiatives. Achieving this mission involves diverse activities spanning sustainability, innovation, outreach, teaching, EDI, and mental health support. BioDocSoc actively participates in school committees, ensuring postdocs' voices are heard and their concerns integrated into school policies. They organize purposeful events, such as the Careers Event, which celebrates diverse career paths and addresses career development challenges. Beyond academics, BioDocSoc promotes the well-being of SBS postdocs, addressing their personal needs. In essence, BioDocSoc is more than a society; it is a vibrant ecosystem nurturing the growth of postdocs and PhD students, bridging the gap between aspirations and accomplishments while benefiting the broader life sciences community in Scotland.

Oral presentations – PhD Students

T6: Spin waves in synthetic antifferomagnets

Jakub Holobrádek,[a] Ondřej Wojewoda,a and Michal Urbánek*[a]

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Synthetic antiferromagnets (SAFs) have become a topic of interest due to their unique properties in the world of magnetics. In this study, we explored how SAFs can control spin waves, which are tiny magnetic vibrations, in a way that might not be immediately obvious.

Think of SAFs like special magnetic sandwiches made of very thin layers. What's fascinating is that by tweaking the layers just right, we can make the spin waves behave in a one-way street manner. Imagine cars driving on a road where they can only go in one direction but not the other. In our SAFs, spin waves move in one direction but not the opposite over a wide range of frequencies.

We achieved this directional spin wave flow by introducing a specific magnetic property called uniaxial anisotropy. It's like giving the road a slight slope that makes cars roll downhill in one direction but not the other. We observed this behavior by interaction of the spin waves with the laser light.

Our SAFs consist of layers that are made very precisely, much like crafting a delicate work of art. We confirmed the quality of these layers using powerful experimental techniques as vibrating sample magnetometry or electron microscope. By understanding how the layers are put together and how they affect spin waves, we can explore new possibilities for controlling spin waves.

Our findings could have implications beyond the world of magnets, potentially leading to new technologies that rely on controlling the flow of energy in one direction, like a one-way valve. This research helps us better understand the hidden properties of materials and how we can use them in exciting ways for future technologies.

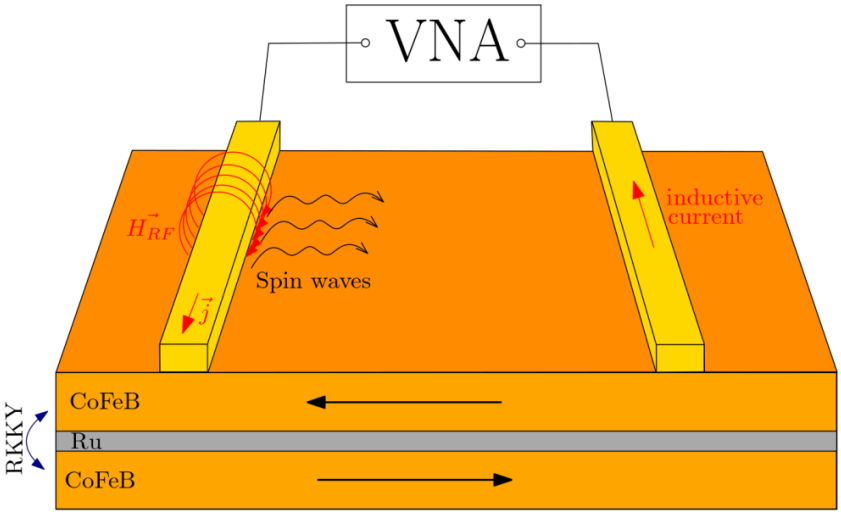

Figure 1: Spin waves measurement in SAF

T7: Distinct p53 Phosphorylation Patterns in Chronic Lymphocytic Leukemia: Where They Come from and How They Affect p53 Function

M. Pesova[a,,b], V. Mancikova[a,b], R. Helma[a,b], S. Pavlova[a,b], V. Hejret[a], P. Taus[a], J. Hynst[a], K. Plevova[a,b,c], J. Kotaskova[a,b,c], J. Malcikova[a, b], S. Pospisilova[a,b,c]

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Protein p53 has a central role in tumor suppression. Under normal conditions, p53 levels are kept low, but in response to DNA damage, p53 becomes stabilized and initiates the transcription of its target genes. In chronic lymphocytic leukemia (CLL), aberrations in the *TP53* gene are associated with the aggressive disease since the function of p53 protein is impaired. However, the p53 function might also be disrupted by other mechanisms, e.g. altered phosphorylation, even in the wild-type protein. Therefore, we aimed to investigate the impact of p53 phosphorylation on p53 function in CLL and uncover what underlies the existence of possible different phosphorylation patterns.

To study this, we induced DNA damage in primary CLL samples with wild-type *TP53* and subsequent electrophoretic analyses followed by western blots revealed two p53 phosphorylation profiles. In profile I samples, p53 was heavily phosphorylated, whereas profile II samples had weakly phosphorylated p53. Whether different phosphorylation patterns affect p53 function to trigger transcription of its target genes was studied by RNA sequencing and validated by qualitative real-time PCR. Profile I samples exhibited a standard response to DNA damage, activating numerous p53 targets. In contrast, profile II failed to fully activate p53 targets, resembling *TP53*-mutated samples. Next, transcriptomic analysis pointed out that untreated cells differed in the basal activity of the hypoxia pathway. The hypoxia pathway was the most activated in *TP53*-mutated samples, profile II was represented by intermediate activity, and the lowest activity was found in profile I. Finally, DNA sequencing revealed that *ATM*, a crucial part of the DNA damage-p53 axis, was more frequently mutated in profile II.

The presented results suggest that lower phosphorylation of p53 in wild-type *TP53* CLL cells results in *TP53* mutant-like behavior regarding the ability to respond to DNA damage. Decreased p53 phosphorylation and related lower ability to respond to DNA damage are linked to genetic defects in *ATM* and the higher basal activity of the hypoxia pathway. Overall, our study highlights the importance of phosphorylation in regulating p53 function in CLL.

This project was supported by GACR 19-15737S, MUNI/A/1224/2022, RVO 65269705, NPO_NUVR_LX22NPO5102.

T8: Discovery of novel HDAC inhibitors for breast cancer treatment using computational approaches

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Histone Deacetylases (HDACs) are the family of epigenetic enzymes that remove acetyl groups from the lysine residues and non-histone proteins. To maintain body homeostasis, enzymes histone acetylases (HATs) and histone deacetylases need to have balance equilibrium. Therefore, any imbalance is associated with tumorigenesis and cancer progression[1]. There are 18 human HDACs isoforms classified into four classes: Class I , II and IV are Zn+ dependent and Class III are NAD+ dependent. HDAC8 belongs to class I member and is reported to be overexpressed in breast cancer initiation and its progression. HDAC8 interacts with the estrogen receptor (ER) and leads to transcriptional inactivation, forming breast cancer[2]. Therefore, HDAC inhibitors are promising therapeutic agents that induce cell cycle arrest and apoptosis of cancer cells. The current study focuses on finding novel HDAC8 inhibitors against breast cancer treatment using molecular modeling approaches. Virtual screening of large library of chemical compounds identifies potential hits that could be drug-like compounds. Further these virtual hits are being filtered using pre-ADMET screening that studies its pharmacological properties. Molecular docking and simulation studies revealed with top virtual hits (NP_1 and EF_1) that have formed a stable complex with the protein with higher binding affinity. MM-GBSA further computed the binding free energy of the top hits and the contribution of individual amino acids that are important in the interaction process. The final hits could be further validated through *in-vitro* experiments to check their viability against breast cancer cell lines.

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T9: An RNA editing-independent function of ADAR1 inhibits PKR activation in mice.

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The deamination of the adenosine base to inosine in dsRNA is catalyzed by the ADAR RNA editing enzymes. In mammals, there are two active ADAR enzymes; ADAR1 and ADAR2. The editing activity of ADAR1 is essential for the discrimination of self and non-self RNA by the innate immune cytoplasmic dsRNA sensors MDA5 and RIGI that signal downstream to MAVS. In humans, mutations in ADAR1 that decrease RNA editing activity cause Aicardi Goutières Syndrome (AGS). Adar null mutant mice are embryonic lethal and die by E12.5. Both conditions are characterized by high aberrant interferon induction. We hypothesized that since Adar, Mavs double mutant mice survive till birth but die as pups, usually within 14 days, other innate immune dsRNA-driven pathways must be in play. Adar null mouse embryos have an increased expression of Pkr (Eif2ak2), a protein kinase that is activated by unedited dsRNA.

We show that the early death of the Adar, Mavs double mutant pups and severe gut defects arising from death of proliferating gut stem cells stem and their aberrant differentiation are rescued in Adar, Mavs, Eif2ak2 triple mutant mice. We also report a regulatory interaction between PKR and ADAR1 that inhibits the phosphorylation of the PKR kinase domain and activation of PKR. Using ADAR1 mutants and deletion proteins expressed in cells, we show that ADAR1 suppresses the activation of PKR through its third dsRNA binding domain (RBDIII) in an editing-independent manner. Our results propose that in addition to dsRNA editing, ADAR1 protein-protein interactions regulate the innate immune response.

T10: Physicochemical properties of tungsten (VI) oxide photoanodes fabricated by wet coating of soluble, particulate and mixed precursors

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Tungsten (VI) oxide coatings on fluorine-doped tin oxide (FTO) glass were fabricated using a wet-coating process with three distinct liquid formulations. The first was based on the dispersion of finely milled WO_3 particles, the second on a fully soluble WO_3 precursor (acetylated peroxotungstic acid), and the third on a mixture of both. The three formulations were deposited by Mayer rod coating onto conductive Fluorine-doped Tin oxide (FTO) substrates. After firing all the three types at 450 °C, WO_3 coatings with significantly different properties were obtained[1]. The fabricated coatings were investigated by profilometry, thermogravimetric analysis, X-ray diffraction (XRD), and Cyclic Voltammetry. [2]The results indicate that the layers obtained from the combination of a particulate ink/soluble precursor (the so-called brick-and-mortar building strategy) exhibit advantageous physicochemical properties, making them well-suited for use as photoanodes in photoelectrochemical cells.

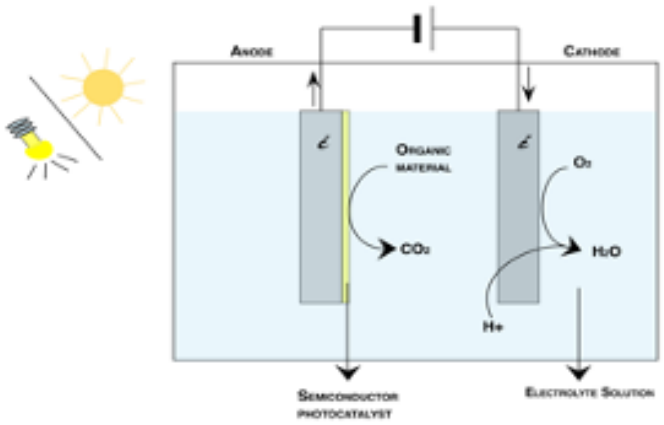

Fig: Schematic of Photoelectrochemical Cell.

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T11: Genomics of Multi-resistant *Escherichia coli* Circulating in a Colony of Gulls: Dynamics of Colonizing Strains Displays Importance of Longitudinal Wildlife Studies

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Transmission of bacteria resistant to antimicrobials in the environment is influenced by humans, domestic animals as well as wild animals. Moreover, antimicrobial resistance genes are spread between bacteria via plasmids. Wild birds, especially wandering and migrating species, are important in this matter since they have the ability to disseminate such bacteria over the entire globe. Our study focuses on the presence of beta-lactam resistant *Escherichia coli* and spread of plasmids carrying resistance genes in the population of wandering wild birds, Caspian gulls (*Larus cachinnans*), breeding at the water reservoir Nové Mlýny, Czech Republic.
During our experiment, five randomly selected non-flying gull nestlings were placed into an aviary for the duration of 12 weeks. Every two weeks, cloacal swabs were obtained from the birds and further cultivated on media with cefotaxime which represented the beta-lactam group of antibiotics. Selected resistant isolates were subjected to whole genome sequencing aiming to analyse their genomic data.
Dynamic changes amongst *E. coli* strains and their transmission between individual birds were observed. In the beginning of the experiment, most of the gulls were colonized by a specific strain of *E. coli* carrying bla_{CMY-2} gene for the production of AmpC beta-lactamases that was located on a F type plasmid. Only one of the isolates carried $bla_{CTX-M-1}$ gene encoding extended-spectrum beta-lactamase on an IncI1 plasmid. During further samplings, we observed that the isolate with F type plasmid disappeared completely and the IncI1 plasmid started to disseminate among various *E. coli* genotypes. At the end of the study, the shedding of the resistant *E. coli* strains in gulls' intestine started to recede. Our results show that resistant bacteria can persist in the gut of migratory birds for a sufficiently long period to be further disseminated into the environment. There is also an exchange of resistant strains between individual birds which leads to a spread of plasmids and might contribute to successful dissemination of new *E. coli* lineages.

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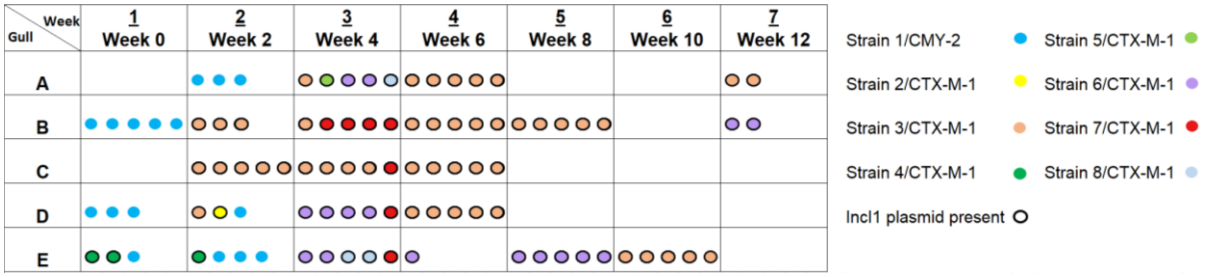

Figure 1. Dissemination of *E. coli* strains among individual birds in time.

T12: Molecular modeling approaches in identification of novel RAGE inhibitors for the treatment of Alzheimer's disease

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Alzheimer's disease (AD) is a progressive neurodegenerative disorder caused by the accumulation of senile plaques composed of amyloid beta (Aβ) protein and neurofibrillary tangles of abnormal hyperphosphorylated tau (τ) protein. The receptor for advanced glycation end products (RAGE) is receptor of the immunoglobulin superfamily that can bind to numerous extracellular and intracellular ligands leading to inflammatory disorders including cancer, diabetes and Alzheimer's disease (AD)[1]. The binding of RAGE to its ligands can trigger various pathways such as *CaMKKβ–AMPK, ERK1/2, GSK-3β and NF-κB* leading to diverse signaling events[2]. Since, RAGE is involved in the pathophysiology of numerous diseases, the targeted inhibition of RAGE or its ligands could be an effective strategy for the treatment of Neurodegenerative disorders including AD. In this study, we used *in-silico* molecular modeling approaches for the identification of novel RAGE inhibitors that serves as antagonists. We expect that the antagonist designed will be able to block the hydrophobic regions on RAGE that interacts with Aβ to prevent the Aβ-RAGE-mediated effects. This in turn will be able to decrease Aβ transport into the brain, increase cerebral flow, decrease neuroinflammation, decrease NF-Kβ activation, decrease levels of proinflammatory cytokines, decrease trafficking of monocytes into brain, decrease BACE1 activation and finally decrease Aβ production.

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T13: Unraveling the Molecular Function of CDM1 Zinc-finger Protein in Meiotic Progression in *Arabidopsis thaliana*

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Meiosis is a special type of cell division in which two rounds of nuclear divisions after one round of genome doubling lead to the formation of four haploid daughter cells. During meiosis, the chromatin is highly condensed, which limits the transcription and suggests that posttranscriptional and translational gene regulation plays a dominant role. Our recent findings indicate that cytoplasmic P-bodies are a central hub for regulating gene expression in Arabidopsis meiosis. *CALLOSE DEFECTIVE- MICROSPORE1(CDM1)* has been described as a transcription factor required to form and dissolve callose in male meiosis (doi: 10.1104/pp.113.233387). However, we found that during meiosis, CDM1 forms distinct cytoplasmic foci that co-localize with DCP1, a marker for P-bodies. CDM1 contains tandem zinc finger motifs, and in-vitro experiments indicated that it has the propensity to form Liquid-Liquid Phase Separation (LLPS) condensates in the presence of RNA. It implies the role of CDM1 in RNA metabolism rather than acting as a transcription factor. To identify RNAs regulated by CDM1, we performed RIP-seq on meiotic tissues and identified several promising candidates. Our further research is aimed at deciphering posttranscriptional role of CDM1 by single molecule RNA Fluorescence in-situ Hybridization (smFISH) method for plant meiotic tissues.

T14: Spectrophotometric Detection of Calcium and Phosphorous in Human Bone Sample, a Straightforward Method in Clinical Analysis

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Inorganic part of human bone mainly consists of hydroxyapatite. The hydroxyapatite component consists of calcium (Ca) and phosphorus (P) elements. Detecting mass ratio of Ca or P to the bone affects the density of bone and can be used to diagnose osteoporosis. Therefore, the simple and accurate methods for determination of Ca and P which do not require costly equipment and skilled operator are highly demanded. Different methods of detection like energy dispersive X-ray spectroscopy (EDX), X-ray fluorescence (XRF), Inductively coupled plasma mass spectroscopy (ICP-MS), atomic absorption spectroscopy (AAS) have been conventionally used for the elemental analysis[1]. However, these methods need expensive instrumentations and trained specialists. Moreover, the mineralization of bone samples is an important step where the mineral content should be fully transferred to the solution phase. This task is usually performed by using a microwave assisted digestion that may introduce another sophistication in instrumentation and operation. Here, in this work, the bone samples are digested in a mixture of formic acid 8% and hydrochloric acid 8%, stirring at 60℃. The Ca^{2+} ion concentration is detected using murexide as a colorimetric reagent that forms a complex with Ca^{2+} ion with an absorption maxima at 498 nm^2. Detection of P is carried out spectrophotometrically by a well-known method using a mixture of potassium antimony tartrate and sodium molybdate as colorimetric reagents at 880 nm^3. A phosphorous to bone mass ratio of (9.8 ±0.47)%, and calcium to bone mass ratio of (16.30±1.29)% are calculated by the described method.

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T15: Guanine Quadruplexes as Targets for the Architectural Protein CTCF

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During interphase, eukaryotic chromatin is arranged into smaller regulatory elements, so-called topologically associated domains (TADs)[1]. These domains are partially involved in the regulation of gene expression by mediating enhancer-promoter interactions within individual domains. TADs are formed by extrusion through the cohesin complex and at their boundaries are anchored by the protein CTCF[3]. The CTCF protein is well known for its ability to bind to DNA and its function as an insulator of gene expression. The eleven zinc finger motifs in the central domain of the CTCF protein indicate its potential to bind to the guanine quadruplex (G4) structure[2]. The recent bioinformatics study confirmed colocalization of the CTCF protein with potential G4 sites within the human genome[4]. G4s are secondary structures of nucleic acids formed by stacking of guanine tetrads, which consists of four guanine residues bound together by Hoogsteen hydrogen bonds. The occurrence of G4s has been confirmed by genome-wide mapping, particularly in the regulatory regions and telomeres[5]. In this study, we have mapped potential G4 motifs of CTCF binding sites within TAD boundaries of the K562 cell line. These regions and their ability to form G4s were then subjected to detailed biophysical analysis, followed by interaction analysis with fragments of recombinant CTCF protein *in vitro*.

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T16: Development of structured substrate for the study of protein curvature sensing on supported lipid membrane

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Curvature sensing in proteins, which is their ability to localize to regions of membranes with specific geometries, underpins fundamental cellular processes, including endocytosis, exocytosis, vesicle trafficking, and cellular signaling[1,2]. Although significant, current techniques for assessing the curvature-sensing ability of peptides are demanding and rely on high-end equipment[3,4]. To address this, we developed a more accessible technique to study protein curvature sensing on lipid membranes of varied geometries. Central to our approach was the use of innovative nanofabrication, notably focused ion beam milling, to craft curved solid supports for lipid bilayers. Guided by preliminary data, we designed specific nanostructures optimal for atomic force microscopy. Our tests confirmed the presence of the lipid bilayer through force spectroscopy and visualization using fluorescently labeled lipids. Using the Zeiss Elyra 7 super-resolution microscope, we identified the labeled peptides' preference for curved membrane regions as characterized by atomic force microscopy. Despite technical and material challenges, our results showcased the efficacy of our nanostructured substrates in studying curvature-sensing peptides. This project, supported by the Jaroslav Koca Bridge Fund, stands as a testament to the synergy of interdisciplinary collaboration, drawing expertise from the CEITEC Nano Core Facility, RoVa group, CEITEC Nanobiotechnology Core Facility, and Cellular Imaging Core Facility.

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T17: Gum Karaya based hydrogels for phage therapy of chronic wounds

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Increasing bacterial resistance impacts wound management of chronic wounds, as it affects over 1 – 2% of people worldwide[1]. Hydrogel materials are widely used to treat chronic wounds, as they provide suitable moisture for wound healing and can transfer active molecules directly into the wound site. This work presents bio-based hydrogel films and gels composed of natural polysaccharide gum Karaya (GK), biocompatible, highly swellable, provides a moist environment, and is potentially antimicrobial[2]. To amplify antimicrobial activity, hydrogels are enriched with bacteriophages, using phage therapy, a promising alternative to treat multidrug-resistant (MDR) bacterial infection as a replacement for conventional antibiotics because phages are a natural enemy of bacteria[3]. This work uses lytic (virulent) phages that infect, replicate, and kill the host cell by lysing the pathogen. Then, the phages are released, and the cycle restarts to eradicate the bacterial colony[4]. This work uses the polyvalent phage JK2 (=812K1/420) of the Myoviridae family, which is an extensive host range of the genus *Staphylococcus,* so it is a suitable candidate for the *Staphylococcus* therapy of chronic wounds[5,6].

The purpose of this work is to develop long-term stable hydrogel materials (freeze-dried films and injectable hydrogels) enriched with phages. Both developed materials proved to be biocompatible and to have sufficient transparency, swelling, and retention behavior and appropriate mechanical properties such as elasticity (up to 120 – 140 % from original size in the hydrated state), easy handling of the film, and optimal injectability (thixotropic character). Phage-enriched GK-hydrogels biostability was tested in three months using the double agar method, which proved phage activity and antibacterial properties against MRSA 1137. These tests proved the biostability of the injectable hydrogel after three months of experiments where the phage reached a concentration above $1 \cdot 10^8$ PFU/ml. Phage visualization in the hydrogel matrix was performed using fluorescent microscopy with labeled phages and GK to determine if they were present in the hydrogel matrix. Based on the results, we can conclude that GK-based hydrogels are a suitable carrier for phages even for long-term storage, treat *S. aureus*-infected chronic wounds, and support the healing process of infected wounds.

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Posters – Postdocs

P1: TiO_2 nanotube Integrated Microwave Resonator UV Sensor

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Ultraviolet (UV) irradiation is extensively utilized in numerous applications such as outer space communication, biological disinfection, memory storage, optoelectronic circuits, and biological analysis [1]. Excessive exposure to UV irradiation is deleterious and causes adverse health effects, for instance, premature aging and skin cancer. A rapid and highly sensitive device for the detection of UV is in great demand in various applications. Recently, planar microwave resonator sensors have demonstrated attractive and robust performance providing high sensitivity, real-time response, and low-cost fabrication process [2]. The planar microwave resonators can easily be integrated with nanostructured materials to make them sensitive to UV radiation via absorption and subsequent charge generation [3]. Among various wide bandgap metal oxides such as TiO_2, ZnO, SnO_2, one-dimensional TiO_2 nanotubes (TNTs) are favorable in UV photodetectors as they possess, except intrinsic TiO_2 properties high active surface area and their unique hollow geometry enables increased charge trapping and, a direct pathway for rapid transport of photogenerated carriers [4-6]. Therefore, the use of high aspect ratio (HAR) TNTs might offers superior sensing performance in the UV region.

In this presentation, the impact of TNT thicknesses on the UV sensitivity of the planar microwave resonator's response will be investigated. We will demonstrate the use of a high frequency microwave resonator integrated with different thicknesses (50, 80, 100 µm) of TNT membranes. The presented work will aid in selecting an optimized thickness of TNT membranes with a large active surface-to-volume ratio to provide the highest sensitivity to UV irradiation. We expect this investigation to act as basis for expanding the use of HAR TNTs to effectively develop low-cost, easy to use and robust microwave UV sensors in a wide range of applications. Experimental details and recent results will be presented and discussed.

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P2: Designer peptides selectively stabilize c MYC quadruplex and propagate apoptosis through c-MYC – VEGF-A – BCL-2 axis in cancer cells

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The pleiotropic proto-oncogene, c-MYC is highly overexpressed in a myriad of cancers and inhabits a transcription-inhibitory quadruplex-forming scaffold (Pu27) upstream P1 promoter. NM23-H2 (Nucleoside diphosphate kinase) unfolds Pu27 to transcriptionally active form while Nucleolin stabilizes the quadruplex thereby restricting transcription. Earlier studies showed a human cathelicidin peptide, LL37, binds weakly to G-quadruplex. Here, we prepared a small library of six peptides by mutating the quadruplex-binding domain of LL37 and investigated their intracellular selectivity for c-MYC quadruplex isomers over other oncogenic quadruplexes by luciferase assays and inspected their magnitude of transcription repression. Then, we analysed the thermodynamic attributes of interactions by calorimetric studies and interpreted in the light of differential non-covalent interactions between peptides and c-MYC quadruplex. We correlated the contribution of interaction by peptide-driven thermal stability, which envisages that the positively charged terminal amino acids make electrostatic interactions with phosphate backbone at 5'-flank and clip the propeller-loop in Pu27. This augments the stability of the quartet favouring cation–pi interactions and pi–pi overlap by internal Trp and Lys residues. We further mapped the stabilized contacts at the atomic level in the solution NMR structure of KR12C–MYC22 complex providing fundamental insights about the positional importance of lysine, leucine, and tryptophan residues to impart higher affinity to the quadruplex by abrogating NM23-H2 recruitment such that even Nucleolin knock-down could not significantly restore cMYC transcription. KR12C promotes apoptotic signalling cascade involving E2F-1–VEGF-A–BCL-2 axis in cancer cells, which opens new avenues towards development of next generation quadruplex-targeting peptides with higher therapeutic-index and minimal off-target effects.

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P3: Cytosine i-motifs containing both DNA and RNA nucleotides

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Cytosine rich regions, which have a potential to form intercalated cytosine tetraplexes (i-motifs, iMs), occur in important parts of genomes, which suggests iM´s role in influencing in various cellular processes. iMs are four stranded DNA structures which assemble at slightly acidic pH and are formed by two parallel duplexes bound together by hemi-protonated cytosine base pairs ($C{\cdot}C^{+}$), which are mutually intercalated in antiparallel fashion. Their properties and occurrence in DNA have been extensively studied, but there is not much known about iM in RNA or about hybrid iM consisted of both DNA and RNA strands. This is in contrast to guanine quadruplexes (G4), which are well studied in DNA and RNA and hybrid DNA/RNA G4 were observed in different sequences in vitro too, including the telomere one, where it could induce exonuclease activity resistance of the telomere end. There is an assumption that this hybrid structure can form even in cytosine strand and possibly it can affect gene expression too.

Formation and stability of iM of sequences with different ratio between DNA and RNA nucleosides was studied using circular dichroism (CD) and UV absorption spectroscopy. Our research shows that stability or even formation of iMs decreases with increasing fraction of RNA nucleosides in cytosine core, but even some DNA/RNA hybrid iMs can form at pH close to the neutral.

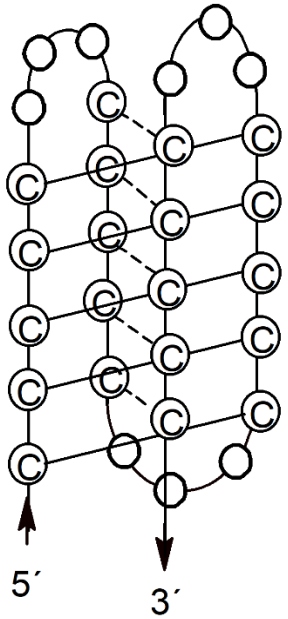

Figure 1. The iM structure of the oligonucleotide with the sequence d(CCCCCTTTCCCCCTTTCCCCCTTTCCCCC).

P4: Electrochemistry of neuromodulation

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Neuromodulation stands at the forefront of medical innovation, revolutionizing the treatment landscape for a myriad of neurological and physiological conditions. This transformative field harnesses the power of electrical stimulation to influence neural circuits, offering new hope and improved quality of life to patients. Current applications span a wide range, from cardiac pacemakers, cochlear implants, deep brain stimulators, to peripheral nerve stimulators, addressing conditions as diverse as cardiac arrhythmias, hearing loss, Parkinson's disease, chronic pain, and more. With ongoing research and development, the potential applications of neuromodulation continue to expand, promising enhanced therapies for an even broader spectrum of conditions.

The foundation of safe and effective neuromodulation lies in the meticulous understanding of the electrochemical processes occurring at the interface between neural tissue and electrodes. These processes are critical for two main reasons. First, they underpin the mechanisms by which electrical stimulation influences neural activity, thereby shaping the therapeutic outcome. Second, they can give rise to harmful electrochemical byproducts that compromise the safety and long-term viability of electrical stimulation.

Our work delves into the intricate electrochemistry of neural stimulation, aiming to elucidate the key reactions that transpire during electrical stimulation. We employ a multi-faceted approach to comprehensively map these reactions, encompassing the direct measurement of electrolysis processes such as oxygen and hydrogen evolution, as well as pH changes. Furthermore, we investigate oxygen reduction reactions[1], quantifying oxygen depletion and the production of hydrogen peroxide. Additionally, we delve into the realm of chlorine chemistry, studying the oxidation of chloride ions to perchlorate ions. Lastly, we explore the dissolution of platinum electrodes in the presence of chloride ions, giving rise to toxic platinum complexes.

In summary, our work sheds light on the intricate electrochemistry underpinning neural stimulation, offering insights into the mechanisms that drive therapeutic outcomes and the potential hazards posed by electrochemical byproducts. We hope that this overview will provide a comprehensive understanding of the critical role electrochemistry plays in the field of neuromodulation, fostering safer and more effective therapies, and inspiring future innovations in this dynamic field.

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P5: Requirements for both Dcr-2 and cGlr1/Sting dsRNA-activated signaling for aberrant innate immune induction in *Adar^{5G1}* null mutant flies lacking adenosine to inosine editing in dsRNA

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Drosophila Adar is mainly expressed in the nervous system [1] and carries out A-to-I RNA editing in dsRNA hairpins in pre-mRNAs. Edited mRNAs are numerous in CNS and enriched in ion channels and neurotransmitter receptor subunits which produce new edited proteoforms (Duan et al. 2017) [2]. Loss of Adar RNA editing activity in *Adar^{5G1}* null mutant flies leads to a severe locomotion defect, consistent with loss of edited CNS proteoforms, and also to aberrant innate immune AMP induction (Deng et al. 2020) [3]. The AMP induction is suppressed by silencing of *Dicer-2* in cholinergic neurons (Deng et al. 2020) [3] and may be similar to aberrant activation of Dicer-related vertebrate cytoplasmic antiviral dsRNA sensors by unedited dsRNA. We sought to determine whether knocking down the antiviral cGas-Like Receptor1 (cGlr1), recently shown to be activated by dsRNA in *Drosophila*, or Sting receptor which acts downstream of cGLR1, can rescue aberrant AMP induction and other defects in *Adar^{5G1}* flies. We found that ubiquitous RNAi knockdown of *cGlr1* in *Adar^{5G1}, arm>cGlr1* RNAi flies rescues the aberrant immune induction in heads; however, it does not rescue the locomotion defect or reduced survival to eclosion. Similar RNAi knockdown of Sting improves survival and rescues aberrant immune induction but not locomotion defects. Furthermore, the double null mutant *Adar^{5G1}; cGlr1 KO* prevents the immune induction and significantly improves the locomotion. It is important to note that *cGLR1 KO* alone has no effect on wildtype locomotion. These data suggest that the innate immunity and neuronal defects in *Adar^{5G1}* null mutant involve both Dcr-2 and cGlr1/Sting pathways, perhaps working together.

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P6: Electron Paramagnetic Resonance studies on Ni(II) complexes

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Molecular compounds are a very flexible platform to modulate magnetic interactions and produce systems such as single-molecular magnets and quantum bits,[1] that have potential for new technologies in data storage and quantum computation.[2] In this work, we describe the preparation of Ni(II) complexes with different ligands intermediating the intermolecular interaction, resulting in features that are observable by electron paramagnetic resonance spectroscopy. Ni(II) is a d^8 transition metal ion with a high spin ground state (S=1) usually presenting a high zero-field splitting (ZFS) and requires high magnetic fields and frequencies to have its intrinsic magnetic parameters correctly characterized.[3] We are reporting on the synthesis and characterization of penta- and hexacoordinated Ni(II) bearing amine or amino acids ligands. The source of Ni^{2+} was the nickel(II) acetate tetrahydrate. All complexes were obtained in moderate and good yields (50%-90%).

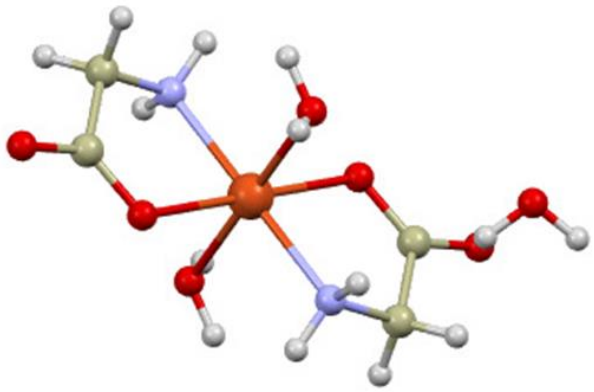

Figure 1. Anticipated hexacoordinated complex of L-histidine with Ni^{2+}.

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P7: Profiling of Adar2 editing in mouse brain

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Glutamate receptors are the primary mediators of excitatory synaptic transmission in the brain. AMPA ionotropic glutamate receptors (AMPARs) are the prime elements that undergo change during synaptic plasticity through alterations in their number, subunit composition, protein partner interactions or phosphorylation state. Gria2 transcript, encoding one of four glutamate receptor subunits of AMPARs (GluA2), undergoes a critical editing event catalyzed by ADAR2. ADARs bind to dsRNA and catalyze a hydrolytic deamination reaction converting adenosine to inosine, which is then read as it were a guanosine by the ribosome, resulting in the insertion of another amino acid. At the Gria2 Q/R editing site a glutamine (Q) codon is converted to an arginine (R) codon. The Q/R editing site is located in the receptor pore loop, so when a positively charged arginine residue is present, AMPARs are resistant to calcium influx while when the glutamine residue is present, then calcium-permeable AMPARs (CP-AMPARs) are produced. This editing event is therefore critical for the correct development of the brain.
ADAR2 mutations in humans cause a very severe, progressive and fatal form of epilepsy that is resistant to treatment with anti-epileptic drugs. Seizures begin during the first year and are associated with developmental regression.
To study how the seizure-prone state progresses, we have dissected the brain from Adar2 mutant pups and their WT siblings at 2 postnatal timepoints before and during the period when they begin to have night seizures. One side of each brain was processed for RNA extraction for studies on expression of Adar2 transcripts and on expression, editing and splicing of transcripts encoding glutamate receptors. The other side was processed for protein samples to study glutamate receptor subunit expression levels with LC MS/MS, followed by confirmational immunoblots against the specific proteins. Our preliminary results show a potential new target of Adar2 editing and several differentially expressed genes potentially involved in epileptogenesis.

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P8: Rapid scan ESR: A versatile tool for the spin relaxation studies at (sub)THz frequencies

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The development of pulse electron spin resonance spectroscopy at microwave frequencies above 100 GHz is rather challenging and expensive task due to the low output power of modern high-frequency solid-state electronics. However, there is a number of scientific problems, e.g., DNP enhancement of NMR, that require spin relaxation measurements at THz frequencies. The rapid scan ESR is an alternative technique that does not require high microwave power and still provides information on the spin relaxation times. The method takes advantage of fast sweeps of the excitation microwave frequency over the ESR line. When the frequency sweep reaches a sufficiently high rate, distinct oscillations (also called wiggles) appear in the spectrum[1-3]. It is possible to retrieve the undistorted (slow-scan) spectrum by employing the Fourier Transform analysis as Josef Dadok had demonstrated in NMR[4]. On the other hand, these oscillations bear information about the electron spin-spin relaxation time, which can be extracted via fitting the rapid scan spectrum using the modified Bloch equations. This technique allows one to capture the spin-spin relaxation time at the nanosecond time scale. Furthermore, the particular design of modern high-frequency ESR spectrometers greatly facilitates the multifrequency operation bringing the spin relaxation measurements to an unprecedentedly broad range of magnetic fields using only one ESR spectrometer (Fig. 1). Finally, we will discuss the future steps necessary to make the THz rapid scan ESR a convenient and easy to use tool for the broad scientific community.

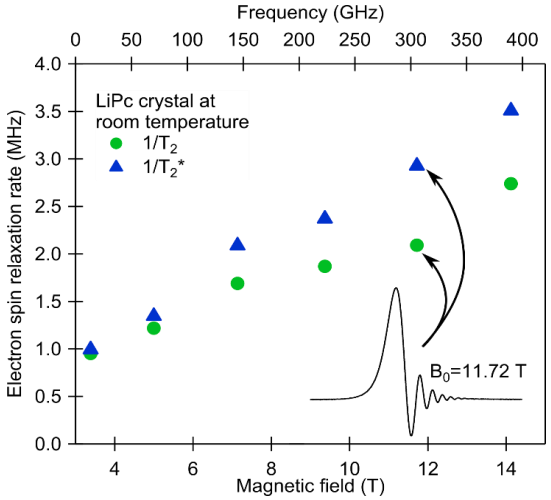

Figure 1. Magnetic field dependence of the electron spin relaxation times T_2 and T_2^* in LiPc at room temperature.

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P9: Manipulation of ADAR1 in Cancer Cell Lines for Neoantigen Discovery

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Recently there has been a huge increase in the number of RNA modifications identified in all types of RNA in all forms of life, currently the number is over 170 modifications. One of the most widespread RNA modifications found in mammals is the deamination of adenosine to inosine in double-stranded (ds)RNA. This process is catalysed by the family of enzymes known as adenosine deaminases acting on RNA (ADARs).
Inosine in dsRNA is recognised as "self" by innate immune RNA sensors [1] whereas non-edited dsRNA molecules are considered as non-self (for instance of viral origin). Unedited dsRNA can lead to the induction of innate immune response. If chronic inflammation is induced it may harm or even kill the cell. In humans, there are two ADAR enzymes: ADAR1 and ADAR2. Mutations in the gene encoding ADAR1 enzymes have been linked to the development of a fatal childhood neurological disease; Aicardi-Goutières syndrome and to a mild skin disorder; Dyschromatosis symmetrica hereditaria.
ADAR1 is induced by interferon and in many types of cancer the level of expression of ADAR1 increases due to inflammation [2]. ADAR1 is an essential gene, so ADAR1 knockdown cause increase levels of unedited dsRNA which leads to the activation of intracellular dsRNA sensors such as melanoma differentiation-associated gene 5 (MDA5) and protein kinase R (PKR) resulting in cell death. We hypothesise that in cancer cell cells, excessive or, on the contrary, decreased amounts of ADAR1 could lead to significant changes in the proteome of the cell which could manifest in the production of neo-peptides, e.g. peptides which are present in the cell only due to changes in RNA editing. If presented on the cell surface, these peptides could attract CD8+ T cells. It has already been shown that peptides generated by RNA editing events can be presented by human leukocyte antigen (HLA) molecules and cause a CD8+ T cell response [3]. Therefore, some of the neo-peptides may be used in the future as biomarkers or as therapeutic vaccines. Thus, the aim of this project is to identify novel neo-antigens that arise to changes in RNA editing.

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P10: Untangling Exact Concentrations of Condensates between RNA Polymerase & RECQL5

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The interaction between RNA polymerase II (Pol II) and transcription factors (TFs) is essential for proper cellular function. The intrinsically disordered regions (IDRs) of TFs have been widely attributed to ensuring their presence at specific stages of transcription, acting as controllers and aids for Pol II's manufacturing of RNA. TFs and Pol II can form condensates, where the interaction between IDRs precisely controls the size and concentration of these molecular hubs[1]. Our understanding of species' spatial organization and concentration within the condensates is limited by the microscopic resolution of densely packed proteins[2]. By characterizing these interactions, the functions and mechanisms of formation in vivo can be more fully understood. One such pair is hyper-phosphorylated Pol II and RECQL5, a TF known to be present during the elongation phase of transcription. Here, we show that cryo-electron tomography can inform molecular dynamics (MD) simulations to make accurate predictions for in vitro condensates. Coarse-grained simulations accurately reproduced the condensate-forming ability of the IDRs of Pol II and RECQL5. Further, the orientation and separation distance of Pol II were matched in silico, estimating the concentration of each species in the condensate in vitro. These results were consistent for condensates containing 10-100 copies of Pol II. Using contact maps from MD simulations, the IDR region of RECQL5, between the Pol II binding KIX domain and the Pol II tail binding SRI domain, is found to be responsible for creating a scaffold that organizes the condensate. Together, the results provide an integrative understanding of Pol II condensate assembly and lay the basis for future studies of more complex cellular condensates using MD informed by tomographic data.

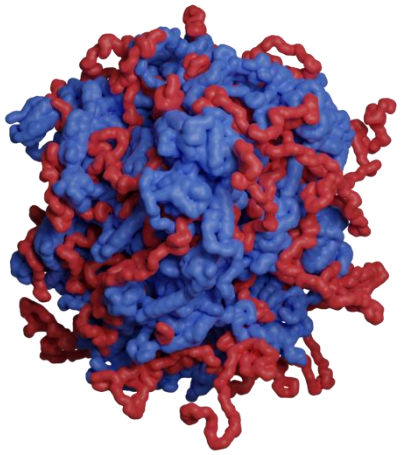

Figure 1. Graphical depiction of a condensate formed of RNA Polymerase II (Blue) and RECQL5 (red). The image is for demonstration purposes only.

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P11: Visualization of tau pathology using in situ cryo-ET

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Misfolded pathologically altered tau protein aggregates into fibrils which further form neurofibrillary tangles that are the hallmark of neurodegeneration in tauopathies, including Alzheimer's disease (AD). Accumulation of misfolded tau in neurons disrupts cellular physiology leading to neuronal death and the propagation of tau misfolding throughout the brain. The presence of pathological tau protein in the brain also affects microglia. They are involved in the uptake and clearance of tau aggregates and the formation of neuroinflammation. Despite the advancements in understanding tau pathology, the relationships between initial tau misfolding, the formation of fibrils, pathology propagation across connected neurons, and subsequent cytotoxicity on the level of individual neurons and microglia remain unclear.

Advances in cryo-electron microscopy techniques now allow studying cellular processes with ultrastructural resolution in the context of whole cells. The possibility of using fluorescent labeling for targeting the area of interest for focus ion beam milling and collecting micrographs or tomograms while working with vitrified cells in a near-native state opened up a new level of resolution.

We use in situ cryo-ET to investigate the ultrastructural aspects of the uptake of AD tau fibrils by neurons and activated microglia. We focus on morphological information about tau neurofibrils and their interactions within neurons focusing on microtubule cytoskeleton and cell trafficking. With microglia, we concentrate on AD tau fibril phagocytosis and how microglia activation affects lysosomes.

In this poster, we present our in situ visualization workflow and show preliminary cryo-ET data of AD-tau fibril intake by microglia.

This work has received funding from the Czech Science Foundation (22-15175I). We acknowledge CF CryoEM of CIISB, Instruct-CZ Centre, supported by MEYS CR (LM2023042)) and European Regional Development Fund-Project „UP CIISB“ (No. CZ.02.1.01/0.0/0.0/18_046/0015974).

P12: New insights into the roles of m6A(m) eraser FTO for cellular homeostasis and DNA repair

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N6-methyladenosine (m6A) and N6,2′-O - dimethyladenosine (m6Am) modifications are present in various classes of RNA within eukaryotic cells. These modifications play crucial roles in multiple aspects of RNA metabolism, including transcription, pre-mRNA processing, localization, stability, and translation. In mammals, the removal of m6A can be carried out by at least two adenosine demethylases: ALKBH5 and FTO. How- ever, the extent of target specificity and functional overlap between FTO and ALKBH5 is currently unknown. Additionally, the regulatory mecha- nisms governing demethylation by each of these erasers remain to be elucidated. In our recent analysis, we employed proximity labeling and mass spectrometry techniques to investigate in cellulo interactions. This analysis uncovered several fascinating factors and protein complexes involved in RNA and DNA metabolism (Covelo et al., 2021). To delve deeper into these discoveries, we conducted protein-protein interaction assays involving FTO and explored the effects of FTO on protein-RNA interactions. Additionally, we conducted a synthetic lethality screen to investigate whether the observed protein interactions with FTO also extend to the genetic level.

P13: Electrophysiology of non-standard model organisms

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The bioelectronics is rapidly advancing field of research, with a growing demand for testing on animal models, primarily favoring mice. Despite its widespread use, this model suffers from disadvantages such as extensive paperwork and inherent complexity. This study seeks to address these limitations by introducing alternative animal models that can be seamlessly integrated into any laboratory setting. These models offer a simplified and easily reproducible approach for the development and testing of bioelectronic devices.
This presentation deals with general implementation of leech and locust models, with a specific focus on their utilization for monitoring electrophysiological responses. We will demonstrate their practical applicability through two bioelectronic devices: the Faraday scalpel and temporal interference stimulation. Through this exploration, we will evaluate the strengths and weaknesses of these models, emphasizing the practical aspects and prerequisites necessary for their implementation in any research facility.

P14: Halfway between pathogens and pollinators. Can bacterial immunity turn bacteriophages into gene transfer agents?

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Bacteria lack recombination as it occurs in eukaryotes. Since recombination is often strongly favoured by natural selection, bacteria have evolved multiple ways to exchange genetic information directly between individuals in a process called horizontal gene transfer (HGT). One of the means of HGT is transfer via gene transfer agents (GTAs), which are bacterial genome-encoded miniscule bacteriophage-derived particles that are released upon environmental trigger and transduce bacterial donor DNA into recipient bacteria, ensuring recombination in the population.[1] Bacteriophage P1 has been shown to be able to mediate generalized transduction between *Escherichia coli* cells.[2] Also, it has been shown to produce dimorphic capsids. The majority are morphologically native bacteriophages, while a small fraction has miniscule capsids.[3] The bacteriophage encodes a *dar* (defense against restriction) operon which ensures the phage DNA is not recognized by three cellular restriction-modification (R-M) immune systems, *Eco*AI, *Eco*BI and *Eco*KI. When the *dar* operon is mutated, the frequency of native to miniscule capsids reverts and most of the capsids are miniscule while a small fraction are native bacteriophages. Simultaneously, the *dar* mutants have increased ability of generalized transduction. The original phenotype is restored on cells lacking *Eco*AI, *Eco*BI and *Eco*KI.[4]

In this project we aim to i) reconstruct the molecular structure of both the native and miniscule phage particles using Cryo-EM, ii) determine how *Eco*AI, *Eco*BI and *Eco*KI products affects phage capsid assembly at the molecular level, and iii) determine whether the miniscule particles are more likely to carry donor DNA than native particles. In other words, we aim to investigate whether bacterial R-M immune systems can turn bacteriophages into GTAs, which would imply that these systems are maintained by natural selection because they allow bacteria to speed up adaptation by recombination.

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P15: Structure of PhiKZ Tail Sheath and Tail Tube

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Bacteriophages, or just phages, are viruses that infect bacteria. With the emergence of antibiotic-resistant bacteria phages emerged as an alternative to antibiotics to treat bacterial infections. *Pseudomonas aeruginosa* is a bacterial species that causes opportunistic infections in hospital patients. The phage phiKZ is a bacteriophage that infects *Pseudomonas aeruginosa*. Here, we present the structure of the tail sheath and tail tube at high resolution. The high-resolution structure enables studying the atomic structure of the tail. It reveals a 6-fold and helical structure similar to other contractile-tailed phages. Each tail sheath protein has extensions that make contacts with 6 other copies, creating a mesh. The void space above and below each protein leave space for the contraction. In the tail tube, the arrangement of the protein in this rigid tube is more compact than in the tail sheath. Besides, each copy has long extensions that intertwine with the neighbouring copies. The structure of both the tail sheath and the tail tube protein is well conserved due to its importance in the phage cycle.

P16: Theoretical and experimental investigation of Fe and Ni-TCNQ on graphene

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Metal 7,7,8,8-tetracyanoquinodimethane (MTCNQ) networks show great promise due to the ready reaction between the organic linker and the transition metal. Therefore, many theoretical and experimental studies have been dedicated to studying MTCNQ-based materials. Although the properties of MTCNQ networks on metal surfaces have been widely reported, still a few studies have considered MTCNQ self-assembly on graphene backbone. In this study, we took advantage of our experimental and theoretical technics to synthesize and reveal the electronic and magnetic properties of Fe and NiTCNQ on graphene as a promising material for diverse applications such as high-density storage media and single-atom catalysis.[1]

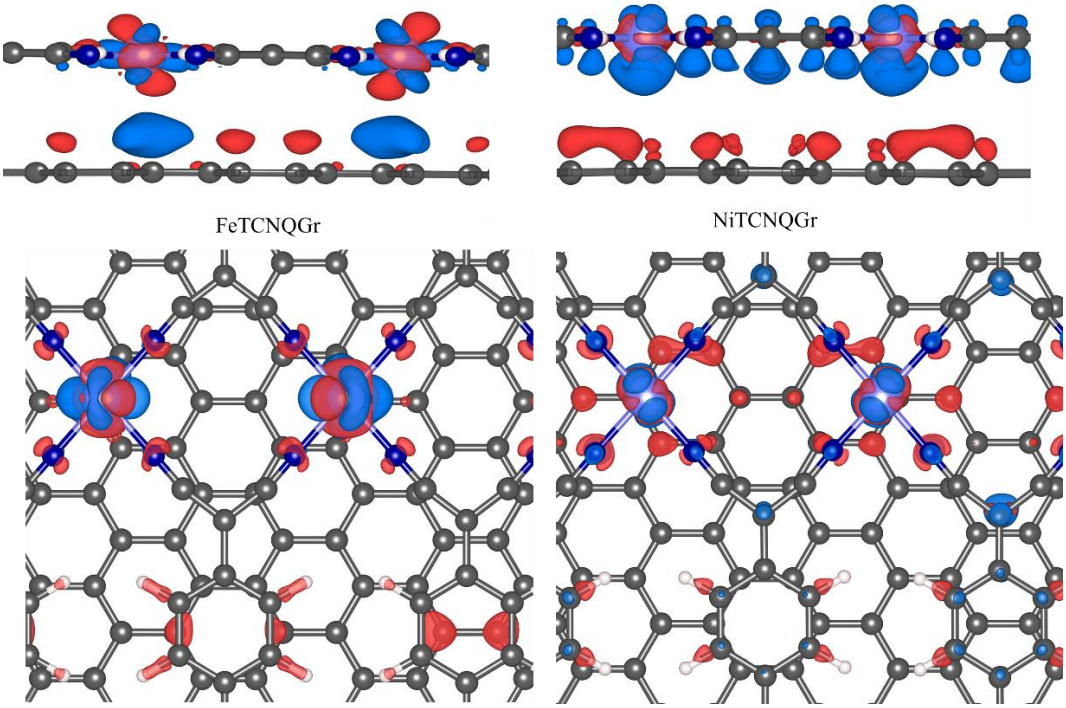

Figure. 1. Charge density difference plot with 0.0003 e/ Å for FeTCNQ/Gr and 0.0004 e/ Å for NiTCNQ/Gr. The blue and red show charge accumulation and depletion distributions, respectively.

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P17: Hidden networks: probing the non-canonical functions of Arabidopsis SMG7

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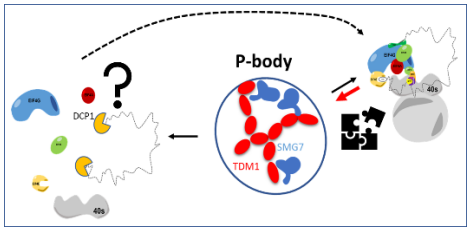
In the life cycle of plants, meiosis marks the transition from the sporophytic to the gametophytic stage. This specialized cell division that produces haploids from diploid cells requires extensive reprogramming of the machinery from mitosis to meiosis and, upon formation of haploid gametes, back to mitosis. Recent studies revealed that during the second meiotic division, a plant germline-specific protein TDM1 is incorporated into Processing bodies (P-bodies) through interaction with the Nonsense-Mediated RNA Decay (NMD) factor SMG7[1]. In addition to its canonical role in NMD, SMG7 perform a crucial function in meiosis germline differentiation in plants, which is distinct from its role in NMD[2,3]. Mutations in either SMG7 or TDM1, lead to third meiotic division and fail to produce pollens, resulting in male sterility. A forward genetic suppressor screen in the background of *smg7* mutants that exhibit reduced fertility was conducted[4], where we identified a mutation in the EVH-1 domain in decapping 1 (DCP1), a critical component of P-bodies.
P-bodies are membrane-less, cytoplasmic ribonucleoprotein granules and are known to be hubs of RNA processing. Control of cellular gene expression via mRNA degradation is crucial to maintain the abundance and life span of cellular mRNA. The DCP1 EVH1-like domain is highly conserved among eukaryotes and is predicted to act like a protein-protein interaction module that recruits specific proteins for decapping, followed by decay. Our phenotypic analysis showed that the *dcp1-4* mutation is among the strongest suppressors of the meiotic defect of *smg7*. This indicates that RNA turnover is a critical determinant of meiotic progression and anther development in plants. Here, we aim to dissect the molecular function of SMG7 and DCP1 during meiosis progression in Arabidopsis.



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P18: Colistin-resistant *Escherichia coli* from Fresh Meat and Slaughtered Animals in the Czech Republic

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Increasing resistance to last-line antimicrobials have been reported from both human and animal sectors. One of these critically important antimicrobials is colistin (polymyxin E), acting on a wide range of Gram-negative bacteria[1]. Whereas colistin usage was seriously limited in human medicine, it was broadly administered in veterinary medicine, especially in poultry and pigs[2]. The aim of this study was to determine the occurrence of plasmid-mediated colistin resistance in domestic and imported meat from retails and slaughter animals in the Czech Republic during 2020–2021 by using selective cultivation and direct PCR testing.

A total of 111 colistin-resistant *Escherichia coli* isolates with *mcr-1* gene were obtained from 65 (9.9%, n = 659) samples and subjected to whole-genome sequencing. Isolates with *mcr* were frequently found in fresh meat from domestic production (14.2%) as well as from import (28.8%). The *mcr-1*-positive *E. coli* isolates predominantly originated from meat samples (16.6%), mainly poultry (27.1%), and only minor part of the isolates came from the cecum (1.7%). In contrast to selective cultivation, 205 (31.1%) samples of whole-community DNA were positive for at least one *mcr* variant, and other genes besides *mcr-1* were detected. Analysis of whole-genome data of sequenced *E. coli* isolates revealed diverse sequence types (STs) including pathogenic lineages and dominance of ST1011 (15.6%) and ST162 (12.8%). Most isolates showed multidrug-resistant profile, and 9% of isolates produced clinically important beta-lactamases. The *mcr-1* gene was predominantly located on one of three conjugative plasmids of IncX4 (83.5%), IncI2 (7.3%), and IncHI2 (7.3%) groups.

The study revealed high occurrence of *mcr* genes in bacteria from fresh meat. Our results confirmed previous assumptions that the livestock, especially poultry production, is an important source of colistin-resistant *E. coli* with the potential of transfer to humans via the food chain. This study brought the first data on dissemination of plasmid-mediated colistin resistance in food production sector in the Czech Republic.

Study was founded by the Internal Grant Agency of the University of Veterinary Sciences Brno, Czech Republic (no. 203/2022/FVHE) and by Czech Science Foundation (no. 22-16786S).

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P19: DNA Methylation Determines Expression of Oncogenic or Tumor Suppressive Isoform of p63 in Squamous Cell Carcinoma

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The *TP63* gene encodes two major isoforms; TAp63 contains a p53-like transcription domain and consequently has tumor suppressor activities whereas ΔNp63 lacks this domain and acts as an oncogene[1]. These two variants are known to show distinct expression patterns in normal tissues and tumors, but the mechanisms involved in their regulation are poorly understood. In squamous epithelial cells with high levels of ΔNp63 and low/undetectable TAp63, the DNA demethylating agent decitabine caused a dose-dependent increase in TAp63, with a simultaneous reduction in ΔNp63, indicating DNA methylation-dependent regulation at the isoform-specific promoters. The reduction was also observed in the direct transcriptional target of ΔNp63, the basal cytokeratin KRT5, which confirms a functional change in p63 activity after DNA demethylation. We also detected high level methylation of three CpG sites in the TAP63 promoter in these cells, which was reduced by decitabine. DNMT1 depletion using inducible shRNAs partially replicated these effects, including an increase in the ratio of TAP63: ΔNP63 mRNAs, a reduction in ΔNp63 protein and reduced KRT5 mRNA levels. We conclude that DNA methylation at the TAP63 promoter normally silences transcription in squamous epithelial cells, indicating DNA methylation as a therapeutic approach to induce this tumor suppressor in cancer. We propose an “either or” mechanism in which TAP63 transcription physically interferes with the ability to initiate transcription from the downstream ΔNP63 promoter on the same DNA strand.

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P20: Phage Phi812 Head Assembly in *Staphylococcus Aureus* Cells

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Opportunistic pathogen *Staphylococcus aureus* causes a broad spectrum of human diseases. Furthermore, antibiotics-resistant and biofilm-forming strains of *S. aureus* contaminate medical devices and cause chronic infections. The lytic phage phi812 from the family *Herelleviridae* has a broad host range, including antibiotics-resistant and biofilm-forming *S. aureus* strains, which makes it a suitable candidate for the treatment of staphylococcal infections. However, it has not been approved for general clinical practice because of the insufficient understanding of many aspects of phage biology.

We inspected phi812 infected cells by cryo-electron microscopy to understand the mechanism of phage assembly. We used focused ion beam milling to prepare electron-transparent lamellas from infected *S. aureus* cells and cryo-electron tomography for three-dimensional reconstructions of the cell content, followed by sub-tomogram averaging of the phage assembly intermediates.

The phage head assembly starts 15 minutes post-infection (m.p.i.) at the inner surface of the cytoplasmic membrane. In a short time (30 m.p.i.), the cell is fully packed with phage assembly intermediates structures, empty, filling, and genome-containing heads and tails connected to fully packed heads. Next, particle coordinates were determined by template matching against a cryo-EM structure of phage phi812K1-420 capsid (EMD-8304). Sub-tomogram averaging of the picked particles revealed distinct classes of phage heads that differ in size and surface features. As many as 313 procapsids averaged in 35 Å resolution, whereas 344 mature capsids gave 32 Å, and the smallest group of genome packaging intermediates heads (152 particles) resolved to 43 Å.

Overall, we described the structure of the previously undescribed procapsid and the phage head in the stage of genome packaging. Our results provide *in situ* structural insight into the phage phi812 head formation in near-native conditions.

P21: Functional Analysis of Small Drug-like Molecules Interacting with mRNA 3'UTRs

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Currently available drugs target only a small portion of disease-related proteins. RNA thus becomes a promising target for small drug-like molecules because, like proteins, it may form pockets ideal for high-affinity interaction with the aforementioned molecules. In order to screen small drug-like compounds, we decided to begin with the 3'-untranslated regions (3'UTRs) of mRNA, which contain highly structured regions. To start with, we chose a few mRNAs, both oncogenic and non-oncogenic, with various 3'UTR structures and lengths. Based on their capacity to bind various RNA structures, we created a library of small drug-like compounds. For an initial screening in-cell analysis using the dual luciferase assay was selected, because it is the most simple, rapid, and sensitive method available. Positive hits from the first round of screening are analyzed in a secondary screening phase by dual luciferase assay together with direct-cell one-step RT-qPCR to examine the RNA integrity in cells upon treatment to small drug-like molecules. With this method in hands, we now are able to perform high-throughput screening of possible 3'UTR binders among small drug-like molecules and choose candidates for further optimization.

Posters – PhD Students

P22: The Role of Long non-coding RNAs in the Microenvironmental Interactions of Malignant B Cells

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Chronic lymphocytic leukemia (CLL) is a disease largely dependent on the interactions of malignant B lymphocytes with the components of the tissue microenvironment (TME). Circulating CLL cells homing to the TME niches in the lymph nodes are provided with stimuli activating B cell receptor (BCR) and critical co-stimulatory signals from other types of immune cells. It has been shown by us and others that microRNAs act as regulators of BCR signaling propensity in the lymph node microenvironment, however, the role of long non-coding RNAs (lncRNAs) in mediating microenvironmental cross talk remains poorly understood.

To identify lncRNAs involved in TME regulation, we analyzed several RNA-seq datasets of differentially expressed lncRNAs and selected a promising lncRNA candidate likely to be associated with the microenvironment. Our data shows that this lncRNA is tightly regulated in response to TME stimuli and BCR-mediated activation of CLL cells, which suggests a regulatory loop fine-tuned by microenvironmental interactions of CLL cells. We determined the lncRNA to be highly expressed in leukemic cells compared to healthy B cells and found it depleted in the expression profile of a panel of blood malignancy cell lines.

Engineered cell line overexpressing the candidate lncRNA was created to probe its biological relevance and molecular function within CLL cells. Extensive phenotyping of the engineered cells in resting cell state didn't show any changes in surface phenotype and phospho-proteomic profile regulating the induction of signaling pathways. RNA-seq profiling of genes differentially expressed in cells with high levels of the candidate lncRNA didn't identify transcriptome pathway fingerprints affecting signaling propensities but showed a mild enrichment of genes involved in cytoskeletal reorganization and membrane trafficking.

We further focused on mimicking the TME by providing relevant stimuli to induce cell activation. Using a functional assay for monitoring intracellular calcium flux, we determined that the lncRNA impairs early-response BCR signaling in activated cells. Additionally, we observed that the lncRNA enhances JAK/STAT6 phosphorylation kinetics in IL-4-activated cells.

The mechanism by which the candidate lncRNA affects these cellular processes is not yet clear. However, we determined the lncRNA to be localized in the cytoplasm, which, taken together with the lack of transcriptional effect, points to the lncRNA's physiological relevance at the signal transduction level. The observed phenotypic changes upon cell activation also suggest that the lncRNA mediates its biological activity on the background of microenvironmentally activated cells, rather than cells in resting state. To elucidate the precise molecular mechanism of action, we are planning to utilize RNA pulldown to map the lncRNA protein interactome.

We identified a novel microenvironment-dependent lncRNA involved in facilitating the cross talk of CLL cells. However, further investigation is needed to understand its precise functional role in the context of microenvironment-dependent regulatory networks.

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P23: Correlative Imaging of Malignant Melanomas Using Laser-Based Techniques, Histology, and Immunohistochemistry

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New methods are being sought to assist histologists in diagnosing tumors. Now, standard excision, the surgical removal of cancerous tissue, including a safety margin of healthy skin, followed by microscopic examination of the specimen, is most used for diagnosis. The problem is the correct estimation of the safety margin during tumor removal. Often incorrect tumor removal of insufficient safety margin leads to the need to repeat the surgery, which can be dangerous, especially for the elderly[1]. Despite that histological examination is the leading diagnostic tool for skin cancer, there is an effort to discover a novel method suitable for preliminary screening.

It has been shown that cancerous tissue changed the shape of the cells and their chemical composition, bioimaging techniques such as LIBS and LA-ICP-MS can be used to image the elemental composition of the tissue, which shows differences in composition of biotic (e.g., C, P, Ca, Mg) and trace (e.g., Zn, Cu) elements between healthy and cancerous tissue[2].

A large sample set of human malignant melanomas was analyzed using LIBS and LA-ICP-MS, and spatial distributions of selected elements were constructed. Immunohistochemical studies were chosen to complete the biological information and confirm the tumor area. The antibody used was Melan A. The aim of this study is to show the potential of correlation of data obtained from all already mentioned methods, which could be used for the possible diagnosis of cancer as a complementary technique to classical histological examination. The next aim is to demonstrate on many malignant melanoma samples that the magnesium, which we can measure in a short time with and with a sufficient spatial resolution by LIBS, could serve as a biomarker for diagnosing skin cancer in the future.

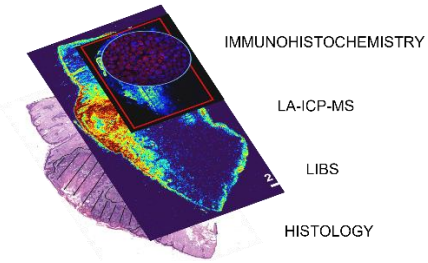

Figure 1. Correlative imaging of tumors using different techniques.

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P24: Dielectric metalens and polarization beam splitter for UV wavelengths

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Manipulation of ultraviolet (UV) light presents a significant challenge for the optical industry not only due to inherent absorption present in the commonly used materials but also due to tight precision limits limiting the fabrication of conventional optical elements such as lenses. Nanophotonics offers a promising solution in the form of metasurfaces, which are able to alter the phase, amplitude, and polarization of incident light[1]. The advantage of metasurfaces lies in their compact size, as well as in the possibility of the implementation of multiple optical functions in a single device[2]. In order to sustain low absorption, which is the key requirement in many optical applications, dielectric Mie-resonant nanostructures are mainly used due to the significantly lower losses compared to the metallic plasmonically active nanostructures[3]. In this work, we choose HfO2 for its high refractive index >2 and wide bandgap 5.7 eV, ensuring low losses down to the deep-UV wavelengths. Based on this material platform, metalenses, metaholograms and self-accelerating beam generators have been demonstrated[4]. In our work, we demonstrate not only a prototypical HfO_2 metalens for UV, but also a polarization beam splitter suitable for wavelengths of 325 nm. In the first step, finite-difference time-domain simulations were performed in order to estimate the phase of the light transmitted by the prospect metasurface building blocks — HfO_2 nanopillars with circular cross-sections on a fused silica substrate which is shown in Figure 1a. Nanostructures of five different diameters were later considered in the metalens design shown in Figure 1b. The subsequent fabrication of the metasurfaces consists of multiple steps, utilizing electron beam lithography into a positive resist and atomic layer deposition. This fabrication process ensures high fabrication precision of the final nanostructures. The design process of the polarization beam splitter, whose function schematic is shown in Figure 1c, is very similar to the one of the metalens. The main difference is in the shape of the nanopillar cross-sections, which had to be rectangular in order to introduce the necessary polarization anisotropy. The metalens and polarization beam splitter are characterized using a custom-made optical setup.

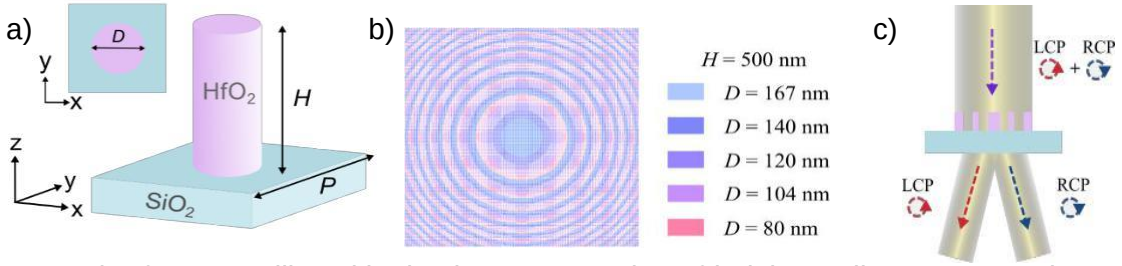

Figure 1: a) HfO_2 nanopillar with circular cross-section of height *H*, diameter *D,* and spacing between the nanopillars *P* on a fused silica substrate. b) Proposed design of the final HfO_2 metalens based on nanopillars 450 nm tall with varying diameters *D*. c) Schematics of polarization beam splitter employing HfO_2 nanopillars with a rectangular cross-section.

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P25: Unlocking Cellular Dimensions: 3D Quantitative non-Invasive Imaging

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Quantitative phase imaging (QPI) is an advanced optical microscopy technique that enables the observation of translucent samples, such as living cells, without any need for fluorescence staining, often associated with photobleaching. Moreover, it offers the unique ability to quantitatively analyze the cells' dry mass distribution, providing invaluable insight when studying processes like cell growth or mitosis.

During the last two decades, a cutting-edge evolution of this technique, known as Holographic Incoherent-light-source Quantitative Phase Imaging (hiQPI), has emerged. Compared to the conventional QPI, the hiQPI method boasts high-quality images with improved lateral resolution and optical sectioning capability[1,2]. As the approach proved to be an essential tool for cancer research[3,4] and drug discovery[5,6], there is an ongoing call to boost the method for a 3D reconstruction modality, a highly sought-after feature in the realm of biomedical imaging. This capability is expected to have an unprecedented impact on cancer research, disease diagnostics, and digital histopathology[7].

The conference poster will introduce a plan to address the 3D reconstruction challenge, the main focus of my Ph.D. studies.

The 3D reconstruction will be based on the so-called Z-stack approach, which utilizes a set of 2D images taken at different axial positions of the sample. Initially, a set of computational phantom cell-like models will be generated. Consequently, these models will be used to simulate the hiQPI Z-stack images, forming the basis for developing robust 3D reconstruction algorithms. Simultaneously, the cell-like phantoms will be manufactured and imaged by an hiQPI-based microscope to obtain an experimental Z-stack. The experimentally acquired images will be compared to the simulated ones and used to optimize the 3D reconstruction algorithms. The outcome of this optimization will be the development of a user-friendly 3D reconstruction software, which will be seamlessly integrated into the hiQPI microscope. In the last phase, the microscope and its new software for 3D quantitative reconstruction will be applied to analyze cells' growth and motility within a volumetric cancer tissue sample.

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P26: Quantitative analysis of cadmium content in tomato leaves by anodic stripping voltammetry at mercury electrode

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Heavy metals may enter the food chain as a result of their absorption by edible plants. Studies have shown that some of these metals have significant toxic and dangerous effects on human health. It is known that excessive exposure to cadmium (Cd) may cause reproductive, skeletal, hepatic, pulmonary, and renal effects and increase the risk of cancer.[1] Therefore, the determination of Cd in environmental samples is very important. Herein we used square wave anodic stripping voltammetry (SWASV) at mercury electrode for ultra-trace determination of Cd in tomato leaves sample. For analysis, the leaves from the control and Cd-treated groups of tomatoes were collected and washed with deionized water. After drying at 100 °C, the samples turned into a powder using a homogenizer and were added to sulfuric acid and hydrogen peroxide and digested two times in a microwave oven. Determination of Cd was carried out in acetate buffer using SWASV at mercury. The plot of peak height against Cd concentration was linear over the concentration range of 5.00-50.0 ppb (R^2 = 0.9978). The detection limit (LOD) and limit of quantification (LOQ) were calculated to be 0.73 ppb and 2.42 ppb, respectively. Moreover, the developed method demonstrated good sensitivity (6.1743 nA/ppb), reproducibility (4.11%), and repeatability (5.90%). In order to control the accuracy of the proposed method, spiked samples (addition of a known amount of standard solution) were measured. The recovery rate, absolute error, and relative standard error were 97.0%, 1.04 ppb, and 2.60%, respectively. The polarographic technique was used to determine the Cd content in digested tomato leaves using the standard addition and calibration curve methods. The content of Cd for the control groups was below the detection limit with both methods. The content of Cd in young and old leaves of the Cd-treated groups was found to be 8.14 ± 0.51 µg/g and 8.02 ± 1.09 µg/g, respectively, using the standard addition method, while they were determined to be 6.78 ± 0.73 µg/g and 7.57 ± 0.81 µg/g using the calibration curve method. The obtained results show that this method is an accurate and reproducible method for the quantitative analysis of cadmium in plants.

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P27: Genetic Predispositions to Hematologic Disorders

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Hematopoietic stem cells (HSCs) produce mature blood cells, such as red blood cells, myeloid cells, lymphoid cells and platelets. Every day 10^{10}-10^{12} new blood cells are formed from the approximately 50 000-200 000 HSCs in an adult human[1]. It was only in 2016 when WHO included predispositions for myeloid malignancies in considerations associated with germline mutations (as an essential component of leukemia diagnosis)[2]. Individuals with germline mutations (predisposition) to hematologic malignancies are diagnosed with increasing frequency. These persons can develop a broad spectrum of hematooncologic disorders (myelodysplastic neoplasia/syndrome, acute leukemia) and also other solid tumors.

Through cell cycle divisions, DNA mutations will over time accumulate. These mutations are known as somatic mutations and will form unique molecular patterns. Most somatic changes will not lead to phenotypic consequences, but some mutations can occur in a portion of the genome giving rise to a relative growth advantage[3]. These mutated HSCs can result in a clonal production of cells derived from a single mutated cell. This cell expansion is referred to as clonal hematopoiesis (CH). As people age, their tissues can accumulate a significant proportion of somatic mutations. CH is highly prevalent in elderly population. Germline and secondary somatic mutations can contribute to final (pathologic) phenotype in individual patient. Recently, results from sequencing analysis have shown that germline variations can influence the developing of CH and HSC function[1,4]. There are only minor prospective data and clinical recommendations for these different patient subgroups and they are usually based on an expert consensus or long-term experience only[5].

The primary objective of the work will be focused on:
A, identification of complex molecular pattern for these patient subgroups
B, correlation it with clinical status
C, proposal of specific ("tailored") recommendations
A broad spectrum of genomic methods will be used and also genetic consulting will be offered.

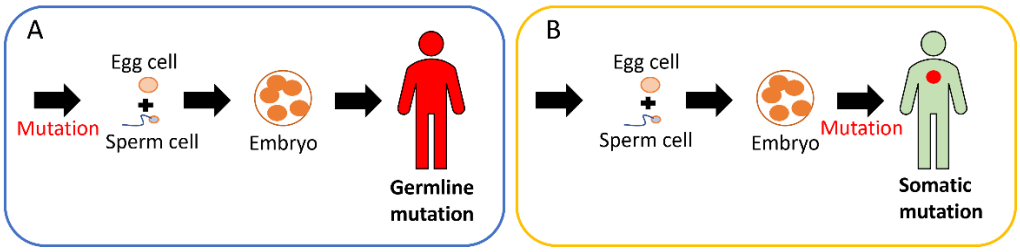

Figure 1. A, Illustration of germline mutation. **B,** Illustration of somatic mutation.

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P28: Can we perceive discrepancies between the senses? A pilot study using virtual reality.

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The sense of body ownership (SBO) refers to the feeling that our body parts and body sensations belong to ourselves. To produce SBO, sensory inputs (visual, tactile, and proprioceptive) must coincide. However, small mismatches among senses are not usually detected and SBO is still induced. Patients with several types of psychiatric disorders (e.g., eating disorders and obsessive-compulsive disorder) show distortions in SBO. We hypothesize that patients suffering from various psychiatric disorders have a different sensitivity to detect multisensory conflicts compared to the healthy population. In a pilot study, we investigated features of the ability to detect a visual-proprioceptive discrepancy in 45 healthy volunteers. For this purpose, we used virtual reality technology. One of the virtual hands was either fixed or shifted outward (max. 20 cm) or inward (max. 10 cm). Participants were instructed to answer whether they perceived a displacement between the virtual and the real hand. According to our results, the ability to detect a visual-proprioceptive conflict depends on the direction of this discrepancy. Participants were more sensitive to inward shifts compared to external shifts of virtual hands. We did not find a significant difference in the ability to detect a multisensory conflict between left and right hands. Next, we will do this experiment on patients with psychiatric disorders. Our paradigm might be efficient in clinical practice as an easy and quick method of detecting a disruption in multisensory integration within mentally ill patients.

P29: Development of a Combined 329 GHz/500 MHz EPR/NMR/DNP System

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Nuclear Magnetic Resonance (NMR), an important analytical method, has found many applications in different research fields.[1] However, despite the tremendous progress made in magnetic resonance techniques over the decades, many biological molecular systems and processes in organisms exhibit so extremely high complexity that investigation of these systems with NMR reaches its limit. The sensitivity of NMR needs to increase further to overcome this limitation, and researchers are actively pursuing novel approaches to enhance the NMR signal.

Dynamic Nuclear Polarization (DNP) stands out as a promising and highly effective technique among the various methods proposed for enhancing NMR sensitivity.[2] DNP increases NMR sensitivity by transferring the spin polarization from electrons via Electron Paramagnetic Resonance (EPR), leading to a significantly larger spin polarization of the nuclei in the studied system.[3] DNP has the potential to revolutionize NMR studies of biological samples, allowing scientists to observe physiological processes in almost real-time.

We present a view on the current state of development of a novel high-field EPR/NMR system operating at a magnetic field of 11.75 T, corresponding to EPR and proton NMR frequencies of 329 GHz and 500 MHz, respectively. The system consists of a custom-built probe capable of measuring ^{1}H, ^{2}D, and ^{13}C NMR and of an EPR bridge featuring a 60 mW solid-state microwave source, suitable for utilization of CW EPR and of a novel Rapid Scan EPR technique developed in our research group. Once finished, this setup will allow us to perform liquid-state DNP, EPR, and NMR experiments in a single system compatible with commercially available NMR instrumentation found in many NMR facilities worldwide.

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P30: Phosphatidylserine-positive Extracellular Vesicles and their Role in Intercellular Communication

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Extracellular vesicles (EVs) are membrane derived particles released by most of the cells of multicellular organisms. Recently, EVs research has attracted much attention, as they are found in all body fluids and their cargo reflects the content of the parental cell. While originally thought of as 'containers' that export waste products from cells to extracellular space, EVs are now known to play an important role in intercellular communication, particularly in the communication of tumor cells with other cells within the tumor microenvironment (TME) [1,2]. Phosphatidylserine (PS) is a phospholipid commonly localized on the inner side of the plasma membrane. During apoptosis, PS translocates to the surface of cells and is found in the membrane of EVs released by apoptotic cells, known as apoptotic bodies. In contrast, viable tumor cells often expose PS on their surface [3]. PS-EVs derived from tumor cells may be critically involved in cell signaling in the TME and in malignant disease progression.

In this work, we isolated PS-EVs from head and neck squamous cell carcinoma (HNSCC) and cancer-derived fibroblasts (CAFs) and analyzed their protein composition. The most abundant proteins were found to be involved in processes such as cell adhesion, angiogenesis, and apoptosis. All these processes critically influence carcinogenesis. PS-EVs derived from CAFs were also used as a 'treatment' of HNSCC cell line, to see their effect on cancer cells. We found that some CAF derived PS-EVs, for example from patient 101, showed resistance to cell death. Therefore, EVs serve as fundamental signal particles with high impact on tumor progression.

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P31: Neutral Cobalt(II)-Bis(Benzimidazole)Pyridine Field-Induced Single-Ion Magnets

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Mononuclear single-molecule magnets (SMMs), also known as single-ion magnets (SIMs), offer a wide range of potential applications, including high-density data storage, quantum computing, and spintronic devices. Unlike materials with long-range magnetic ordering, the magnetic bistability of SMMs is solely based on molecular properties and does not depend on intermolecular interactions [1]**Chyba! Nenalezen zdroj odkazů.**.
Four novel pentacoordinate Co(II)-based field indiced single-ion magnets were prepared and characterised. The complexes feature the metal centre coordinated with one two terminal bromido or chlorido ligands along with tridentate derivatives of 2,6-bis(1*H*-benzimidazole-2-yl)pyridine ligand containing either n-octyl or n-dodecyl chains. The presence of long aliphatic chains ensures the solubility in the solvents frequently used for lithography patterning, such as chloroform.
The processability of compounds was tested by lithographically controlled wetting and a versatile wet-lithography.

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P32: Liposome Encapsulation of Calendula Officinalis Extract for Controlled Release in DFU Treatment

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Diabetes affects millions globally, leading to diabetic foot ulcers (DFUs)[1] with impaired healing and bacterial biofilm formation. Due to the misuse of antibiotics[2], there is a need for non-antibiotic alternatives, such as *Calendula Officinalis* extract, supporting faster wound closure and greater size reduction compared to antibiotics[3], improving the DFU treatment. Encapsulation into liposomes preserves the antioxidative and antibacterial activity of the extract, while FUR hydrogel creates a drug-delivery system with controllable degradation, mechanical properties, and antibacterial activity for the intended application. In our previous study, the *Calendula* extract was found to be unstable. To preserve its activity, increase bioavailability, and ensure controlled release, the extract was encapsulated into liposomes using the Mozafari method (*n=5*). The liposomes remained stable over 28 days, maintaining diameters of 85.7 ± 3.9 nm and a consistent zeta potential -18.9 ± 2.1 mV. Encapsulation efficiency, antioxidant activity, and total phenolic content were measured, showing positive effects and confirming the suitability of the encapsulated extract for the intended application. The liposomal *Calendula Officinalis* extract will be further incorporated into a suitable hydrogel and the release will be evaluated.

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P33: Diazepam Effects in *Arabidopsis Thaliana*

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Key words: benzodiazepines, *Arabidopsis Thaliana*, plant communication, stress

Diazepam is one of the most prevalent anxiolytic drugs, consumed by patients worldwide on a large scale. In mammals, it binds to benzodiazepine binding site of GABA receptor and inhibits excitation. In plants, GABA inhibits anion passage through Aluminium-activated malate transporter family of proteins (ALMTs) and therefore plays a role in modulation of plant growth, development, and stress responses.[1, 2] We have investigated the influence of diazepam on *Arabidopsis Thaliana* growth, germination, seed production and gene expression.

Experiments evaluating germination, primary root length, hypocotyl and seed production were performed using stratified *A. Thaliana* Col-0 seeds that were sown on diazepam spiked MS media plates (10 mg/L). Seeds yield experiments were performed by spraying *A. Thaliana* with diazepam solution throughout whole life cycle. Experiments focusing on gene expression used *A. Thaliana* DR5, GCamP3, JAZ10, MYC2, pCYCD6, R-GECO1, VSP2 and WER lines and roots were evaluated by confocal microscopy in two parallel sets – plants wounded by a tweezer and non-wounded controls.

Results show no impact on germination rates and hypocotyl, however, treated plants have shorter primary roots and yield significantly higher number of seeds. In terms of confocal microscopy, DR5, GCamP3 and pCYCD6 were unaffected in both wounded and non-wounded plants. JAZ10, MYC2 and WER were induced only in wounded plants while VSP2 was induced only in non-wounded plants. R-GECO1 was supressed only in non-wounded samples.

Further molecular studies are needed to assess the interaction pathways. Diazepam appears to intersect both Ca^{2+} and jasmonate signalling pathways and we hypothesize that there is a connection with in-plant GABA-like receptors which may even contain benzodiazepine-specific binding site. This research implies parallels between molecular mode of action in both animals and plants and can reveal the broad effects of diazepam-like compounds on stability, diversity, and health of natural ecosystems.

Research was carried out in collaboration with Peter Marhavy's research group at Umeå Plant Science Center, Sweden.

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P34: Wastewaters as a Source of Multi-drug Resistant *Klebsiella* spp. for Environment

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Antimicrobial resistance is one of the most serious threats to global public health. It causes longer hospitalization and higher treatment costs, as well as increased mortality. Antibiotic-resistant bacteria are found not only in humans and animals, but also in food and the environment[1]. Municipal wastewater treatment plants (mWWTPs) have been identified as a hotspot facilitating the spread of antimicrobial resistance into the environment. *Klebsiella* spp. is frequently associated with nosocomial infections and have the potential to spread outside hospitals via wastewaters. Modern molecular methods including whole genome sequencing allow detailed analysis and comparison of bacterial isolates from different sources. Sequencing and bioinformatic analyses can provide detailed information on mechanisms of resistance, virulence and mobile genetic elements and their ability to spread[2]. The aim of this study was to track the spread of multi-drug resistant *Klebsiella* spp. from the clinic to the environment via wastewaters.
In total 383 *Klebsiella* spp. isolates were obtained from patients and five types of waters (hospital sewage, inflow and outflow from mWWTP, river upstream and downstream mWWTP) from three cities in the Czech Republic. All isolates were tested for susceptibility to 24 antibiotics by determination of the minimum inhibitory concentration, production of extended-spectrum beta-lactamases (ESBL) and were subjected to whole genome sequencing.
Most isolates (95%) showed multi-drug resistant profile with predominant resistance to beta-lactam antibiotics (93%), sulfonamides (85%), ciprofloxacin (87%) and tetracycline (66%). Some isolates showed resistance to last line antibiotics carbapenems (14%) and colistin (8%). ESBL production was confirmed in 97% of isolates and was predominantly encoded by $bla_{\text{CTX-M-15}}$ gene. Carbapenemase-encoding genes were found in isolates originating from all sample types including river. Isolates were assigned to 78 different sequence types (STs) with predominance of clinically relevant *K. pneumoniae* ST307 and ST405 found in all water types and clinical samples. Clonal transmission from clinical isolates via mWWTP to the river water was confirmed for ST307 strains.
This study confirmed the ability of highly virulent antibiotic-resistant *Klebsiella* spp. to spread from healthcare settings to the environment. Presence of strain resistant to last line antibiotics in the surface water highlights the importance of monitoring the environment for the presence of clinically relevant bacteria.

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P35: Recognition of RNA Polymerase II C-terminal domain by RPRD2

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The largest subunit of human RNA Polymerase II (RNAPII) contains highly flexible C-terminal domain (CTD) that is composed of 52 heptapeptide repeats (first half of repeats with consensus sequence YSPTSPS and second half largely degenerated in sequence). Several CTDs canonical and non-canonical residues can be subjects of post-translational modifications. Tyrosine, threonine, and serine residues undergo dynamic phosphorylation/dephosphorylation resulting in specific phosphorylation patterns throughout different stages of transcription cycle. These phosphorylation patterns are recognized by various transcription and processing factors during the transcription cycle. Therefore, CTD plays an important role in the regulation of transcription and coupling of transcription to post-transcriptional processes such as mRNA processing.

One of the human transcription factors that recognizes phosphorylated RNAPII CTD is RPRD2, but its exact role in the transcription cycle is still poorly understood. In this study, we show that RPRD2 recognizes specifically pSer2 or pThr4 phosphorylated forms of CTD via its CTD-interacting domain (CID). The interaction of RPRD2 CID with pSer2 phosphorylated CTD is further enhanced by additional phosphorylation on pSer7. To provide mechanistic details of the interaction between RPRD2 CID with pSer2,7 CTD or pThr4 CTD, we solved the solution structures of both complexes using NMR spectroscopy. RNAPII CTD pSer2 and pThr4 phosphorylation occur during the late elongation and termination in yeast and humans. RPRD2 preference for these phospho-marks shown in this study, and similarity at some aspects to yeast transcription termination factor Rtt103, suggests the possible involvement of RPRD2 in late elongation or transcription termination.

P36: Encapsulation and Biological Studies of Bioactive Ruthenium(III) Complexes in CD-MOF-1

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Ruthenium-DMSO complexes are promising among ruthenium-based anticancer drugs due to their good selectivity for solid tumor metastases and minimal host toxicity. Substantial improvement over earlier Ru-DMSO anticancer complexes occurred with NAMI and NAMI-A* compounds. Nevertheless, given the poor stability, low water solubility, and yet some toxicity associated with the unspecific delivery of these metallodrugs, research has focused on the encapsulation of ruthenium complexes into host systems.[1] With the rapid advancement of material chemistry, much effort has been directed to the formation of innovative platforms for controlled and smart drug release systems with the goal of maximizing therapeutic efficacy while minimizing side effects. One of such systems are Metal-Organic Frameworks (MOFs), composed of organic ligands and metal ions/clusters linked together by coordinative bonds forming 1, 2, or 3-D networks.[2] Cyclodextrins (CDs) are hydrophilic cyclic oligosaccharides having truncated cone-shaped molecules. The outside hydrophilic surface is decorated by a number of hydroxyl groups, whereas the inner hydrophobic cavities are lined by glycosidic oxygens and C-H units. The γ-CD (8 glucose unit) has been utilized in green synthesis of biocompatible and non-toxic MOFs (CD-MOFs) using alkali and alkaline earth metal ions.[3] In this research the model complex (MC) composed of ruthenium(III) metal center coordinated with 4-methylpyridine and DMSO as axial ligands and four chlorides in equatorial plane was synthesized, studied for its stability, and used for adsorption studies into CD-MOF-1 (composed of potassium hydroxide and γ-CD). Herein, we present the first pharmaceutical formulation of potential metallodrug inside the CD-MOF-1 and it's *in vitro* viability studies on spheroids of human hepatoblastoma cell line HepG2 (Figure 1).

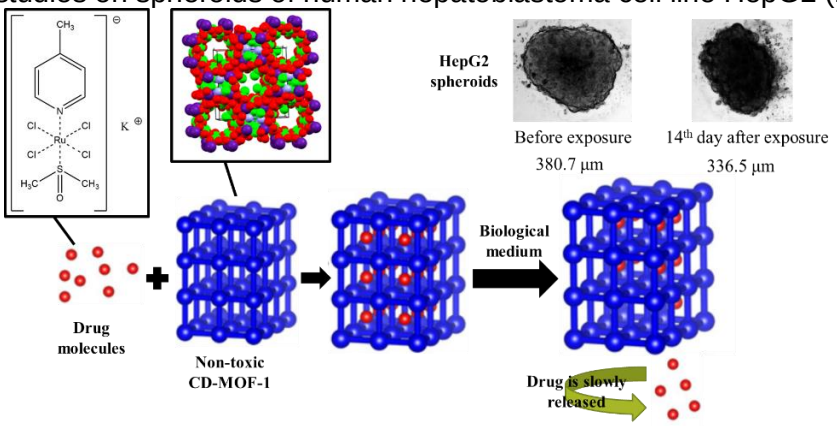

Figure 1. Graphical abstract

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P37: Genome-wide CRISPR/Cas9 knockout screening revealed genes involved in CD20 regulation in rituximab-resistant cells

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CD20 antigen has been used as a target of monoclonal antibodies (mAb) such as rituximab (RTX) in the therapy of B-cell malignancies for more than two decades. However, malignant B cells downregulate CD20 on their surface, resulting in mAb resistance and therapy failure. Therefore, it is crucial to investigate the CD20 regulation to enhance the efficacy of antiCD20 mAb. This project aimed to perform CRISPR/Cas9 knockout screening to identify genes whose disruption restores CD20 surface expression.

To create a model mimicking the situation in patients who have developed resistance to mAb therapy, we generated RTX-resistant CD20-low B-cell line by chronic exposure to rituximab. These cells were transduced by the GeCKO lentiviral library to obtain a collection of single-gene knockouts. After 2.5-week cultivation, the top 5% of cells with the highest expression of CD20 were sorted out. Using next-generation sequencing, we identified gene knockouts responsible for CD20 upregulation.

CRISPR/Cas9 screening revealed several genes whose disruption increased CD20 surface expression. CSK, encoding a negative regulator of Src kinases, as well as PTEN, a well-known tumor suppressor, were among the top hits. These two genes are involved in the B-cell receptor (BCR) pathway – an essential pathway in B cells. Interestingly, we identified four genes SSR1- 4, encoding all four subunits of the TRAP complex, an endoplasmic reticular complex involved in protein translocation across ER membrane. STT3A, encoding the catalytic subunit of oligosaccharyltransferase, was another ER-associated gene revealed by the screening. These results indicate that both BCR signalling and ER play an important role in CD20 regulation. Selected genes were validated, and the mechanism of their function is being investigated. The understanding of underlying mechanisms could provide a way for a potential enhancement of anti-CD20 mAb therapy.

P38: MAGNETIC RESONANCE SPECTROSCOPY of THIN MOLECULAR FILMS

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Molecular magnetism has been attracting the attention of scientists from diverse disciplines because of its potential applications in spintronic devices, quantum bits (qubits), and high-density information storage. Molecular magnetic compounds offer the flexibility of playing with different organic ligands around a single or multiple spin centres for obtaining a desired magnetic performance. The interplay between the magnetic and structural properties is essential for the design of new and effective materials with a specific application, such as Single-Molecule Magnets (SMM), molecules that present a slow relaxation of magnetization for data storage. High-Field Electron Paramagnetic Resonance (HFEPR) and Fourier-transformed Infrared Magnetic Spectroscopy (FIRMS) are powerful approaches to reveal magneto-structural parameters enabling the determination of the magnetic anisotropy, g-tensor values, and electron spin relaxation mechanisms.[1] Aiming at the potential application of these systems in devices, their ability to be deposited intact on surfaces, the characterization of the resulting thin-films, and the interaction with the substrates is a necessary step. We will present recent achievements of our research group that allow the sublimation of SMM on substrates using a high vacuum chamber for the production of thin films, the characterization of bulk and thin-film materials using our home-made sample holders for HFEPR spectroscopy,[2] and a coupling mechanism of an FTIR spectrometer to our superconducting magnet allowing the measurements of FIRMS. Besides that, we will present other important techniques to evaluate the surface chemistry of the prepared thin-films, like scanning electron microscopy, X-ray photoelectron spectroscopy, and atomic force microscopy.

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P39: Deciphering Tick-Borne Encephalitis Virus Neutralisation by Monoclonal Antibodies using cryo-EM

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Tick-borne encephalitis (TBE) is a serious illness caused by the tick-borne encephalitis virus (TBEV), known to induce potentially fatal inflammation in the central nervous system. Despite the availability of vaccines, regions heavily impacted by TBE, like the Czech Republic, continue to exhibit low vaccination rates[1]. The **increasing incidence of TBE** cases underscores the urgency for targeted therapeutic interventions. While **antibodies have exhibited promise as a treatment** modality in a mouse model, involving intravenous administration of TBEV-specific antibodies, our comprehension of the underlying molecular mechanisms governing TBEV neutralisation remains limited[2].

Our study delved into the intricate interactions between the **TBEV Neudörfl strain** and two specific neutralising mouse monoclonal antibodies: IC3 and A4, which **target distinct domains of the TBEV envelope protein**[3]. The investigation was carried out using isolated TBEV from infected tissue culture cells, subsequently combining it with Fab fragments derived from the neutralising antibodies. These mixtures were then vitrified on electron microscopy grids, enabling cryo-electron microscopy analysis. Following data collection, single-particle analysis was used to elucidate the structural configurations of the TBEV-Fab complexes.

Exploring the molecular foundation of TBEV neutralisation by antibodies could enhance our comprehension of the significance of diverse epitopes located on the viral surface. This knowledge might pave the way for the customised development of therapeutic antibodies or the creation of more precise vaccines in the years ahead.

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P40: INVESTIGATING THE MOLECULAR MECHANISM OF THE PIVOTAL AUXIN SIGNALING COMPONENT AUXIN RESPONSE FACTOR 5 IN EMBRYOGENEIC TRANSITION IN *ARABIDOPSIS THALIANA*

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In *Arabidopsis thaliana*, the *in vitro* culture of immature zygotic embryos at a late stage of development on a solid medium containing synthetic auxin 2,4-D leads to the formation of somatic embryos via direct somatic embryogenesis (SE). It exemplifies the unique capacity of pluripotency of somatic cells. This developmental process provides an appealing model system for studying embryogenesis and understanding the molecular mechanisms controlling totipotency and pluripotency in plant somatic cells. It is widely exploited to regenerate plants *in vitro*, for plant mass micropropagation, for the protection of plant biodiversity, and for the production of transgenic plants.

Previous research and preliminary results suggest that the phytohormone auxin via one of the key auxin response factors, AUXIN RESPONSE FACTOR 5 (ARF5), is essential for SE induction, most probably by controlling genes involved in auxin production (YUCCA) and signaling (MIR390). Those regulators play a significant role in SE by regulating auxin homeostasis during the embryogenic reprogramming of Arabidopsis somatic cells.

The project aims to unravel the nature of the regulatory relationship between ARF5, the auxin production via the transcriptional regulation of *YUCCA* genes, and auxin signaling via MIR390 in controlling the SE process based on the use of various methodologies, including CRISPR-induced mutagenesis and plant transformation, MIR390 sensors design and development, somatic embryogenesis induction, dual-luciferase reporter assays, hormonal profiling, gene expression analysis, greenCUT&RUN method, etc.

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P41: The Antibacterial Ability of Novel Robocast Ti-Cu Scaffolds

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Combating implant-related infections is a major challenge for the current healthcare systems since bacteria demonstrate an ultimate resistance to antibiotics and defense of host tissues stimulating the inflammation and frequently persisting until the implant removal[1-3]. This has driven the development of new sophisticated implantable materials with intrinsic antibacterial properties as an alternative or additional measure to prevent infections. In this work, Ti-xCu antibacterial scaffolds were designed and fabricated for the very first time by robocasting and *in situ* alloying during sintering. The scaffolds were sintered under two different regimens[4] to study the effects of Cu content (x = 0, 3, 5 wt. %) and sintering remaining porosity on the antibacterial activity *in vitro*. The Ti-Cu scaffolds developed a hypoeutectoid microstructure of α-Ti(Cu) + Ti_2Cu, confirmed by XRD and EDX analyses. Ti-Cu alloys sintered for a shorter time had smaller grain size and open porosity that resulted respectively in higher strength and strain of the scaffolds. In addition, the hypoeutectoid microstructure together with the remaining porosity significantly decreased the corrosion resistance of the scaffolds and rendered their contact-killing antibacterial activity. The Ti-3Cu-P scaffold was the strongest and had the highest antibacterial activity against *S. aureus*. *In vitro* culture of Saos-2 cells confirmed good cytocompatibility of Ti-Cu scaffolds. In conclusion, robocasting offers a unique opportunity to use Ti-Cu powder mixtures as feedstock to fabricate antibacterial Ti-Cu alloy *in situ* with a potential use in the reconstruction of skeletal defects and as bone tissue engineering scaffolds that minimize the chances to develop implant related infections.

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P42: The Role of Adar in Regulation of Innate Immunity in *Drosophila*

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Adenosine deaminases acting on RNA (ADARs) are enzymes that deaminate adenosine (A) to inosine (I) in double-stranded RNA (dsRNA) structures. In vertebrates, loss of ADAR1 RNA editing leads to aberrant activation of interferon and interferon-stimulated genes by activation of antiviral cytoplasmic dsRNA sensors which recognize endogenous dsRNA as virus-derived. Drosophila lacks these cytoplasmic antiviral dsRNA sensors and their closest relative in in Drosophila is Dicer2. There is only one *Adar* gene in *Drosophila melanogaster* and *Adar*^{5G1} null mutants show aberrant innate immune induction of antimicrobial peptide (AMP) transcripts that requires the dsRNA sensor Dicer-2.[1] *Drosophila* Adar also carries out efficient site-specific RNA editing in CNS transcripts to recode over six hundred proteins; this is unrelated to innate immunity.

The aim of this study is investigation of the pathway leading to induction of AMP transcripts in *Adar*^{5G1} null mutants by identifying further mutations that can suppress the aberrant innate immune induction. To achieve this, we use the *UAS/Gal4* binary system, specifically a weak ubiquitous (*Arm-GAL4*) driver, to knock-down different components of the Dcr-2 innate immune signaling pathway. Our preliminary results show rescue of AMPs levels in *Adar*^{5G1} null mutants by knocking down *Dcr-2* or *r2d2*, one of Dcr-2's main cofactors. In addition, it seems that overexpression of human ADAR1 p150 isoform is sufficient to prevent aberrant AMP transcript induction in *Adar*^{5G1} null mutants.

In parallel, we knocked out *Adar* in Schneider S2 cells, derived from larval hemocytes, and checked levels of immune induction in comparison to wild-type S2 cells. *Adar* knock-out lines do not show an increased immune induction compared to wild type in normal conditions. Also, neither *Adar* knock-out nor normal S2 cells show increased immune induction in response to synthetic dsRNA transfected into the cytoplasm. We observe a complete lack of expression in S2 cells of the *cGLR1* transcript encoding the main sensor of dsRNA.[2] Work on the cGLR1/Sting pathway in *Adar*^{5G1} mutant flies, presented in a poster by Khadijja Hajji, shows that both Dcr2 and Sting signaling are involved in aberrant innate immune induction in *Adar*^{5G1} mutant flies.

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P43: Employment of 5D NUS NMR experiment for disordered protein with repetitive motives assignment

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Nuclear Magnetic resonance (NMR) is often the method of choice in structural biology when characterizing intrinsically disordered regions (IDRs). However, when using standard triple resonance assignment experiments the successful IDR assignment often becomes unfeasible, as IDRs signal strongly overlaps due to frequent repetitive motifs and numerous prolines. ^{13}C-detected 5D non-uniformly sampled (NUS) experiments overcome these challenges in assigning IDRs.[1,2] This experiment enabled to assign over 99% of Tau residues successfully. The assignment efficiency is noteworthy as the 2N4R Tau variant contains 18 aa stretches of the motifs VXSK to PGGG and a proline-rich domain. The assignment provided information about secondary structure propensities and proline-conformation analysis. The obtained assignment will be used to determine amino acids essential for interaction with binding partners e.g., 14-3-3 proteins. Moreover, this approach was recently employed for the IDR (residues 38-137) of the extracellular surface variant of carbonic anhydrase IX (CA IXsv, residues 38-391) associated with aggressive tumor growth and metastasis.[3] As CA IX´s activity is increased with the presence of IDR and lower extracellular pH typical for solid tumors, the obtained assignment will be subsequently utilized to investigate the influence of various pH conditions on its local structure features and involvement in the activity of CA IX.[4,5]

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P44: Effects of Solvent Media on the Electrochemical Activity of $Ti_3C_2T_x$ MXene Electrocatalysts

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Hydrogen has been proposed as a clean and promising energy carrier that supports the development of renewable energy sources in order to overcome the global warming crisis. Therefore, the focus is on the generation of "green" hydrogen via the electrolysis of water. As a one-half reaction in the water-splitting process, the hydrogen evolution reaction (HER) is considered a promising approach for clean hydrogen production. Platinum-based electrocatalysts are found to be the most efficient for the HER. However, their scarcity and high cost represent the biggest obstacles in their large-scale use. Thus, developing non-noble metal-based catalysts for the HER has attracted a lot of attention.

A fast-growing family of two-dimensional materials, MXenes, has drawn much attention due to their excellent catalytic properties and wide range of applications.[1] One of the most common ways to work with powder-based materials, such as $Ti_3C_2T_x$ MXenes, consists in the dispersion of the catalytic materials in various solvents followed by drop-casting of the prepared suspensions onto desired surfaces.[2] However, the solvent employed to prepare the powder dispersions can have noticeable effects on the electrochemical performance of the drop-cast samples. Herein, four different solvents (water, ethanol, isopropanol, and N,N′-dimethylformamide), used as dispersion media in the preparation of the $Ti_3C_2T_x$ MXene samples, and their effects on the electrocatalytic activity of $Ti_3C_2T_x$ MXenes towards the HER were studied by linear sweep voltammetry. As shown in this work, the different solvents provoke changes in the roughness and dispersibility of the material, as well as a partial oxidation of the $Ti_3C_2T_x$ microparticles which leads to dissimilar surface coverages, resulting in different overpotential values for the catalytic HER.[3] The findings of this research are of crucial importance for knowledge about the electrocatalytic performance of MXene samples, and their storage and long-term usage.

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P45: Detection and Investigation of Oxidatively Damaged RNA

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Oxidative stress plays a major role in biological systems occurring naturally in physiological processes.[1,2] In particular, significant amounts of reactive oxygen species (ROS) are produced in mitochondria as part of the respiratory chain. On the other hand, the immune system generates hypochlorous acid (HOCl), catalyzed by the enzyme myeloperoxidase, to fight pathogens as part of the immune response. HOCl induces chlorination and oxidation of various biomolecules such as lipids, peptides and nucleic acids, altering their native function and structure.[3] Known modifications include *N*-chloramines, 8-chloro- and 8-oxopurines as well as 5-chloro- and 5-oxopyrimidines. Especially, guanine as the most susceptible nucleobase towards oxidation has been the primary target of investigation, leading to the identification of several oxidized derivatives such as spiroiminodihydantoin (Sp), guanidinohydantoin (Gh), oxazolone (Z), imidazolone (Iz) and diimino-imidazole (Diz). Oxidation-induced damage has been well investigated on the level of DNA; however, the effect on RNA is poorly understood.

Our goal is to investigate the effects of oxidative stress on RNA using methods such as UV absorbance, LC-MS/MS, and Next-Generation Sequencing (NGS). Furthermore, a novel NGS method is established named OxiAbSeq, a chemistry-based deep sequencing method to study RNA oxidation profiles, in particular G oxidation products and abasic sites.[4,5]

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P46: Self-sorting of Platinum(II) Complexes Inside Cucurbit[8]uril

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Introduction of cisplatin as a chemotherapeutic agent significantly improved the survival rate for many cancer patients. Following the discovery of cisplatin, numerous cisplatin-like compounds have been introduced worldwide but only two compounds, oxaliplatin and carboplatin have been approved by FDA. Despite the great success of cisplatin and its analogous bifunctional compounds, these compounds have several side effects due to low selectivity toward cancer cells and binding to the off-target biological nucleophiles.[1] To overcome the challenges several thousand drug candidates have been reported in the past few decades but they are not void of similar drawbacks. The concept of supramolecular prodrug can be used to make the drug protected and more selective towards tumor cells. In this approach, the drug is entrapped into a carrier molecule thereby achieving temporary deactivation and extending its lifetime in the extracellular environment, enabling the anti-tumor warhead to reach its target efficiently.[2]

In this contribution, we will demonstrate new supramolecular drug-carrier systems based on monofunctional Platinum(II) complexes and cucurbit[8]uril (CB8). We designed new monofunctional Pt(II) compounds with aromatic anchors and characterized their binding to CB8. Due to the larger size of CB8 cavity, it can encapsulate two of the Pt(II) guests. We synthesized two types of ternary assemblies, i) homo-ternary assembly (two same Pt(II) complex), ii) hetero-ternary assembly (two different Pt(II) complex) inside CB8. Time-dependent ^{1}H-NMR reveals the presence of several kinetically stable forms depending on the orientation of the two Pt(II) guests and the hydrolyzed state of the Pt(II) center, but after several days those converge to one thermodynamically stable form. 2D-ROESY spectra and mass spectrometry revealed a very unusual head-head orientation of two highly charged Pt(II) centers (**Fig 1**).

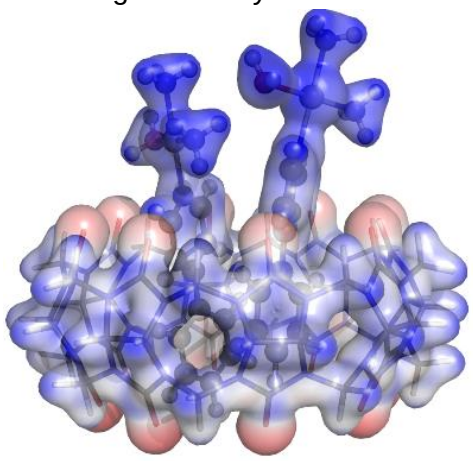

Fig 1: ESP map of homo-ternary assembly

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P47: A Novel Approach to Chemical Derivatization of Pseudouridine to Improve RNAseq-based Detection

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Pseudouridine is the most abundant modified nucleoside in RNA and was first discovered in 1951. Pseudouridine displays similar characteristics to uridine, such as identical Watson-Crick base pairing with adenine and an identical molecular mass. In order to distinguish these two bases, liquid chromatography (LC) in combination with downstream mass spectrometry (MS) can be used, making use of differing elution times and fragmentation patterns. As most information about the sequence context are lost during LC-MS analysis, further sequence-specific analysis is necessary to retrieve site-specific pseudouridine quantities in a distinct RNA.[1] Hence, several methods for sequencing of pseudouridine have been developed, such as Pseudo-seq, Ψ-seq, PSI-seq and CeU-seq, all relying on selective chemical treatment with CMCT.[2] Chemical modification can also be found in nanopore sequencing, such as labeling inosine with acrylonitrile as shown in Nano ICE-Seq.[3] While chemical treatments with CMCT, acrylonitrile and methyl vinyl sulfone are commonly used, these bear several limitations such as incomplete reactions with pseudouridine and side reactions with other nucleotides.[1] Therefore, in the context of 'RMaP' project C01, other Michael acceptors were tested at different reaction conditions in order to detect pseudouridine with a high specificity and a preferably high sensitivity to enable mapping of pseudouridine at single nucleotide resolution. Reaction efficiency and side product formation of different reagents were monitored via LC-MS of digested tRNA after chemical treatment and will be followed by extensive testing on several artificial and native RNAs containing pseudouridine. Propargyl acrylate (PA) appears to be a good candidate with high reactivity, which also allows further derivatization by click chemistry and ester hydrolysis. Finally, biological samples will be treated with promising candidates and analyzed via Illumina and Oxford Nanopore sequencing to elaborate efficiency and applicability of our new approach.

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P48: Effects of water on the static and cyclic compressive strength of the 3D-printed hydroxyapatite scaffolds

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The environment of the human body is complex, and it differs in many aspects from that in a laboratory or other dry setting. Bone implants are usually exposed to different bodily fluids, such as blood, interstitial fluid, and synovial fluid, which can affect the mechanical behavior and implant integration with surrounding tissues. Therefore, testing of bone scaffolds in wet conditions before the implantation is important to ensure their safety and efficacy in human body. This work focuses on the mechanical testing of hydroxyapatite scaffolds and composites in watered conditions and the comparison of the results against the dry setting. Hydroxyapatite scaffolds were produced by 3D printing, and were mechanically reinforced with a hydroxyapatite foam, producing a hydroxyapatite/hydroxyapatite composite. Infiltrated and blank scaffolds were tested under static and cyclic compression. The results showed that reinforcement of the porous hydroxyapatite scaffold with the ceramic foam improved the strength of the scaffold approximately two times. It was observed that water considerably reduced the static compressive strength by up to 50 %, showing a significant weakening of the hydroxyapatite structure, when exposed to the liquid. However, water had quite opposite effect during the cyclic compression, where water improved the survival of the structures and resistance to the block cyclic loading. In conclusion, liquid conditions can noticeably affect the mechanical behavior of hydroxyapatite scaffolds and composites, with water being significantly detrimental for the compressive strength. This work was supported by the BUT project CEITEC-VUT/FCH-J-23-8367.

P49: *In situ* rapid analysis of interferon gamma

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Interferon-gamma belongs to the group of antiviral cytokines which has been known to have a very important role in the human defense system. It is also particularly responsible for providing adequate ability for the body to fight against important diseases such as autoimmune diseases, cancer, AIDs, and tuberculosis. Moreover, it can be used as a biomarker to see if there is contamination of some diseases such as tuberculosis.[1] Therefore, it is very important to rapidly, cost-effectively, user-friendly, and portably obtain the accurate and precise amount of interferon-gamma in human blood for therapeutic, pharmaceutical, etc. purposes. Some patients need to be monitored in real-time. thus, the need to equip the detection with up-to-date data transmission systems is felt very much. Currently, there are methods to determine this value, but they have limitations such as expensiveness, not being portable, requiring an expert, and slow detection time without the ability to analyze and transfer data using the Internet of Things to a medical center. Therefore, in the present study, the aim is to design an electronics-compatible system for in situ detecting interferon-gamma in human blood using a biosensor.

P50: Electric-Field Control of Qubits in Quantum Paraelectrics

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The application of ultrabroad band HFERP is a promising technique to investigate the influence of an electric field on the electronic and magnetic properties of quantum materials. The coexistence of electric and magnetic degrees of freedom in paraelectric materials with spins makes the electric-field manipulation of spins possible [1]. In this perspective, high field, and high frequency EPR offer higher sensitivity, higher g-value resolution and access to larger zero-field splitting of spin energy levels in broad frequency range (85-1100GHz). The interplay between different HFERP techniques namely continuous wave and pulsed EPR measurements will allow the study of spin transitions induced by the electric field component of microwave and spin-electric field coupling in quantum paraelectric materials. Pulsed EPR measurement is more convenient at low temperature where the relaxation time is longer, however, measurement of high dielectric permittivity materials is challenging and require special treatment for microwaves field distribution in the resonator cavity. Spin dynamics was recently demonstrated by means of Rapid Scan HFEPR a new developed technique where sweep rate up to 300000 THZ/s was achieved in the range of 170-250 GHz [2-4]. Hence, making rapid scan EPR more advantageous for lesser resonator cavity experiments. In this poster we discuss the feasibility of HFEPR techniques: continuous wave HFEPR, and multi-frequency rapid scan HFEPR as well as pulsed ERP for high dielectric permittivity materials under electric field, their characteristics, limitations, and challenges will be presented as well.

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P51: Advanced Bone Regeneration: 3D Printed Scaffolds Enhanced with Bioactive Proteins

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Extensive bone defects pose a significant research challenge thanks to their slow healing process. The application of 3D printing offers a compelling alternative to traditional methods involving manual processing and casting. This is primarily due to its capacity to define the shape, dimensions, and structure of the product. This revolutionary technique facilitates the precise, and cost-efficient production of bone substitutes for regenerative medicine.
In this project, a unique bio-ink was prepared by mixing calcium phosphate ceramic powder (CaP) with a thixotropic biodegradable thermosensitive copolymer and a biocompatible polysaccharide. The main goal was to create a non-toxic and completely bioresorbable material suitable for the human body. This bio-ink exhibits thixotropic characteristics, enabling it to transition into a printable biomaterial suitable for direct ink writing (DIW) technology. The scaffolds were carefully crafted following a biomimetic approach involving 3D printing at lower temperatures.
To support the healing process, bioactive proteins with the ability to boost regeneration or suppressing infections were infused into the printed scaffolds. These proteins were incorporated after the printing process using a coating technique. Subsequently, the scaffolds were solidified in a high-humidity environment. The release of proteins was conducted at body temperature within a physiological solution (phosphate buffer saline) and monitored using enzyme-linked immunosorbent assay (ELISA). The structural and morphological characteristics of the printed samples, printed at varying extrusion coefficients, were analyzed using scanning electron microscopy. This comprehensive analysis offered valuable insights into the structural and morphological alterations induced by distinct printing conditions.

This work was supported by the ProfiBONE project (TO01000309) that benefits from the EEA Grants and the TACR within the KAPPA Programme. CzechNanoLab project LM2023051 funded by MEYS CR is gratefully acknowledged for the financial support of the measurements at CEITEC Nano Research Infrastructure.

P52: Functional Integration of a Semi-synthetic Azido-queuosine Derivative into Translation and a tRNA Modification Circuit

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Supplementation of an azide-functionalized synthetic analogue of preQ1, a precursor of the queuine nucleobase (q), resulted in efficient incorporation into tRNA in vitro and in vivo in Escherichia coli (E. coli) and Schizosaccharomyces pombe (S. pombe). The azide moiety enabled for click chemistry allowing for analytical applications through click conjugation with a fluorescent dye or biotin for affinity purification. The incorporated analogue was shown to be present on actively translating ribosomes, indicating a functional integration into aminoacylation and ribosomal recruitment. Furthermore, the semi-synthetic tRNA modification, here termed Q-L1, also functionally replaced the naturally occurring queuosine modification (Q) in tRNA maturation by mimicking its stimulating effect on the methylation of C38 in tRNAAsp. This demonstrates the functional integration of a synthetic clickable moiety into a modification circuit, where one RNA modification stimulates another. In conclusion, the scarce queuosinylation sites in cellular RNA makes the synthetic q/Q system a minimally invasive method to introduce non-natural nucleobases.

P53: Molecular Architects of the Genome: Cooperation of Guanine Quadruplexes and CCCTC-Binding Factor on Chromatin Organization

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DNA molecules are key part of the cell nucleus. Their secondary structures are essential in various cellular processes and chromatin three-dimensional architecture.[1] Interphase chromosomes are dissected into chromosome territories, organized into areas of up to a million base pairs in size – topologically associated domains (TADs) and subdomains. Border areas of topologically associated domains are highly enriched in non-canonical tetra-stranded secondary structures of nucleic acids, guanine quadruplexes (G4s). Guanine quadruplexes are established by stacking of square planar structures, guanine tetrads, mainly in the presence of K+ ions. Four guanine bases present in guanine tetrads are associated through Hoogsteen hydrogen bonding. The formation of topologically associated domains is also mediated by architectural proteins, like the CCCTC-binding factor (CTCF) and cohesin complex proteins – SMC1, SMC3, and RAD21.[2,3] Protein CTCF generally serves as a transcriptional insulator by blocking communication between enhancers and promotors. Architectural protein occupancy strongly corresponds to the content of G-quadruplexes formed at TADs boundaries. The bond between CTCF and DNA is mediated by 11 zinc-finger motifs, which were predicted to interact with G4s.[4] This study aims to biophysically characterize the topology and stability of G-quadruplexes at the TAD boundaries of the K562 cell line. The second part of this study is to understand the interaction between guanine quadruplexes and transcriptional repressor CTCF, both in vitro and in cells.

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P54: Synthesis and swelling behavior of metal-chelating superabsorbent hydrogels based on sodium alginate-g-poly(AMPS-co-AA-co-AM) obtained under microwave irradiation

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A metal-chelating superabsorbent hydrogel based on poly(2-acrylamido- 2-methylpropanesulfonic acid-co-acrylic acid-co-acrylamide) grafted onto sodium alginate backbone, NaAlg-g-poly(AMPS-co-AA-co-AM) is prepared under microwave irradiation. The Taguchi method is used for the optimization of synthetic parameters of the hydrogel based on water absorbency. The Taguchi L_9 (3^4) orthogonal array is chosen for experimental design. Mass concentrations of crosslinker MBA C_{MBA} initiator KPS C_{KPS}, sodium alginate C_{NaAlg} and mass ratio of monomers $C_{AM/AA/AMPS}$ are chosen as four factors. The analysis of variance of the test results indicates the following optimal conditions: 0.8 g L^{-1} of MBA, 0.9 g L^{-1} of KPS, 8 g L^{-1} of NaAlg and $R_{AM/AA/AMPS}$ equals to 1:1.1:1.1. The maximum water absorbency of the optimized final hydrogel is found to be 822 g g^{-1}. The relative thermal stability of the optimized hydrogel in comparison with sodium alginate is demonstrated via thermogravimetric analysis. The prepared hydrogel is characterized by FTIR spectroscopy and scanning electron microscopy. The influence of the environmental parameters on water absorbency such as the pH and the ionic force is also investigated. The optimized hydrogel is used as adsorbent for hazardous heavy metal ions Pb(II), Cd(II), Ni(II) and Cu(II) and their competitive adsorption is also discussed. Isotherm of adsorption and effect of pH, adsorption dosage and recyclability are investigated. The results show that the maximum adsorption capacities of lead and cadmium ions on the hydrogel are 628.93 and 456.62 mg g^{-1}, respectively. The adsorption is well described by Langmuir isotherm model. The hydrogel is also utilized for the loading of potassium nitrate as an active agrochemical agent and the release of this active agent has also been investigated.

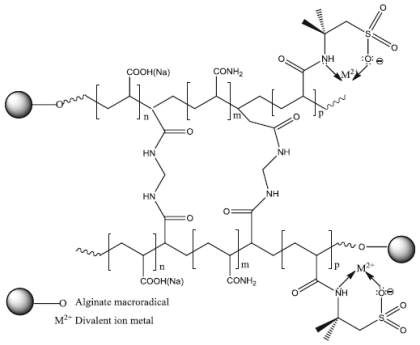

Figure 1. Chelating role of AMPS in the hydrogel.

P55: ph-Responsive Microcapsules Loaded with Copper Nanoparticles for the Treatment of Infected Burn Wounds

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Infected burns and chronic wounds can escalate into huge complications, including the potentially life-threatening condition of sepsis. In numerous cases, traditionally used antibiotics have been ineffective, because of the resistance exhibited by multiple bacterial strains. Notably, there has been a growing focus on the development of new materials, enriched with inorganic nanoparticles, to be effective against bacterial strains that have acquired resistance to antibiotics. Over recent years, numerous drug and nanoparticle delivery systems have been developed, featuring pH-triggered release tailored to the acidic pH environment for cancer treatment. Surprisingly, there have been only a few studies that aimed to form pH-responsive delivery systems for release in alkaline pH region of infectious wounds. In this project, our primary goal was to synthesize antibacterial copper nanoparticles (CuNPs) that are encapsulated in poly(lactic acid) capsules with faster release in alkaline pH region of the infectious wound. The faster release is based on the fundamental principle of electrostatic repulsion. The synthesis of copper nanoparticles starts with chemical reduction, followed by stabilization with chitosan of both, low (LMw) and medium molecular weight (MMw). Stabilized nanoparticles were encapsulated in pH-responsive capsules made of poly(lactic acid), a biodegradable polyacid widely used in medicine. Prepared samples were characterized by transmission electron microscopy, dynamic light scattering, and UV-VIS spectroscopy. Our preliminary results from CuNPs synthesis involved the comparison of LMw and MMw chitosan as stabilizing agents. CuNPs coated with MMw chitosan stabilized particles had an average radius of 203.9 nm, while those coated with LMw chitosan exhibited an average radius of 170.4 nm. It's noteworthy that LMw chitosan-coated particles exhibited a reduced tendency to form clusters, a characteristic that may be advantageous in subsequent encapsulation. In conclusion, many different materials, such as scaffolds, wound dressings, or hydrogels can be enriched with these antibacterial particles and used for wound infection treatment.

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P56: Investigating the novel mutations in *ADARB1* effect its activity

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In recent years, we have witnessed the emergence of a burgeoning field known as epitranscriptomics. One of the most abundant and extensively studied RNA modifications within this field is the deamination of adenosine to inosine by the family of enzyme; adenosine deaminase acting on RNA (ADAR). Three members of the ADAR family, ADAR1, ADAR2, and ADAR3, have been identified in humans, but only ADAR1 and ADAR2 are enzymatically active. ADAR2 plays a key role in regulating neuronal excitability and inhibition. It is involved in editing of transcripts encoding ion channels and neurotransmitter receptors, proteins critical for neuronal electrical signalling[1]. ADAR2's editing activity can modulate the function of these proteins, thereby impacting on the excitability levels of neurons.

Previous research has shown that some ADAR2 mutations found in children with seizures can reduce RNA editing efficiency, which may be related to severe symptoms[2]. Therefore, understanding ADAR2's functions and the consequences of its dysregulation is of significant interest for both basic neuroscience research and the development of potential therapies for related diseases.

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P57: The Effect of Phosphoserine on Degradation and Mechanical Properties of Composite Bone Adhesive

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Composite bone cement, widely used in orthopaedics and dentistry, often exhibits limitations in its mechanical properties. This study focuses on the modification of bone cement with phosphoserine and its impact on the mechanical properties and degradation behaviour of modified bone cement. Phosphoserine, naturally found in the human body and linked to bone mineralization, was chosen as the additive. The modified cement preparation was optimized through additive adjustments, ratios, and integration methods. Mechanical tests, including compression tests, and X-ray diffraction analysis were used to measure cement properties and chemical composition. The results of the mechanical tests show significant improvements, with a maximum strength of 50% enhancement compared to the unmodified cement. Degradation tests confirmed higher weight loss with more phosphoserine, and scanning electron microscopy provided surface insights before and after degradation.
Introducing phosphoserine as an additive enhances the mechanical strength and degradation characteristics of composite bone cement. This discovery has great potential to improve bone cement performance and longevity in orthopaedics, dentistry, and bone-implant advancements.

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P58: Photofaradaic microstructures for photostimulation

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Hydrogen peroxide (H_2O_2) is a reactive oxygen species involved in many biological pathways. H_2O_2 act as signaling molecule in low nM concentration range, while exposure to higher levels of H_2O_2 can lead to cytotoxicity[1]. The aim of this work is to create injectable photosensitive microparticles, that would produce physiological amounts of hydrogen peroxide to regulate signaling pathways *in vitro* or *in vivo*. The proposed microparticles are substrate-free metal/semiconductor particles with a thickness of less than 100 nm and a micrometer size in other dimensions. Production of H_2O_2 is determined by photofaradaic oxygen reduction reaction with concurrent oxidation reaction of an electron donor molecule present in the electrolyte[2]. The photofaradaic reactions are driven by continuous exposure to red light (650 nm) which gives the possibility of transferring the technology *in vivo*. The poster will detail the fabrication process of the microparticles and discuss the characterization of H2O2 production using spectrophotometry assay.

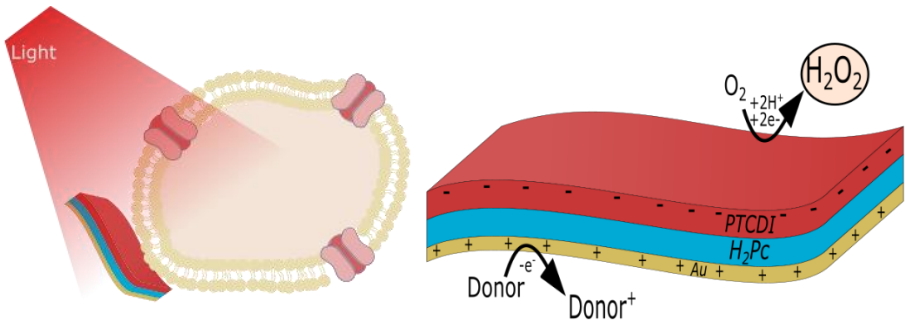

Figure 1. The production of hydrogen peroxide with concurrent oxidation reaction driven by red light source

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P59: Targeting RNA with Small Molecules: A Therapeutic Frontier

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Many drugs currently available target proteins, but around 80% of those proteins are considered undruggable; therefore, selecting another suitable candidate is crucial for treating many systemic and cancer-related pathologies. Around 75 % of the human genome is transcribed into RNA, while only a small fraction (～3%) of it translates to protein. Hence, RNA can be exploited as a potential therapeutic target. Specifically, the 3' untranslated regions (3'UTRs) of RNAs which play a crucial role in mRNA stability and gene expression regulation, have distinct secondary and tertiary structures that interact with proteins within cells. In this study, the goal is to identify small drug-like molecules that can target the 3'UTRs of several non-druggable oncogenes and non-oncogene addiction genes, such as MYC, KRAS, HSF1, CDK12, NRF2, etc.

A library of specialized heterocyclic compounds representing drug-like small molecules was screened against 120 nucleotide fragments of the MYC mRNA 3'UTR using a high-throughput fluorescence-based anisotropy assay (FA). Several fragments showed significant changes in fluorescence anisotropy when mixed with small molecules, indicating potential binding and conformational changes in the RNA structures. Selected RNA fragments were subsequently screened using Surface Plasmon Resonance (SPR), a highly sensitive biophysical technique where RNA is immobilized, and small molecules are flowed across the surface. Significant binders were identified, indicating the potential of these small molecules to interact with the RNA targets.

Our screening approach leverages the conformational changes in RNA fragments upon small molecule binding. Further characterization of these interactions will enable us to create a library of RNA motif-small molecule interaction pairs that can be utilized to target other mRNAs. These interaction pairs will aid in identifying lead compounds capable of modulating mRNAs and mRNA-protein complexes within cells. Disrupting RNA-protein interactions with small molecules can result in the downregulation of disease-related protein expression, with potential therapeutic implications.

P60: CRISPR/Cas9 Technology as a Useful Tool in the Study of Chronic Lymphocytic Leukemia

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Chronic lymphocytic leukemia (CLL) is a heterogeneous disease with various somatic mutations, the most frequent of which targeting *ATM*, *TP53*, *NOTCH1*, *MYD88* and *SF3B1* genes. Their thorough exploration could shed light on the disease etiology, or even lead to discovery of potential novel drug targets. However, patient CLL cells do not proliferate *ex vivo*, thus precluding lengthy experiments, such as CRISPR/Cas9 screening.

Using CRISPR/Cas9 in CLL-derived HG3 and MEC1 cells, we generated isogenic cell lines carrying disruptive mutations in *ATM* or *TP53*. These cell lines show complete loss of the respective proteins and abrogation of downstream signaling pathways. We also used CRISPR/Cas9-based homology directed repair to obtain HG3 cells with recurrent mutations of *NOTCH1* (P2514fs), *SF3B1* (K700E) and *MYD88* (L265P).

Selected cell lines were subjected to CRISPR/Cas9 dropout screening to identify genes, whose deletion is lethal to the introduced mutations. In particular, *SPDYE1* and *LUC7L3* were found to be synthetically lethal with the *NOTCH1* mutation, while *SNUPN* and *UQCRC1* were found to be essential for *SF3B1*-mutated cells. Simultaneously, the cell lines were screened with a library of 859 approved drugs. The screening demonstrated sensitivity of *NOTCH1*-mutant and *SF3B1*-mutant cell lines towards inhibitors of various hormone receptors or inhibitors of 20S proteasome. The knockout models were also used for studies of the performance of anti-CD19 CAR T-cells. We observed different effectiveness at eradicating tumor cells in vivo depending on the driver mutation, with *TP53* mutations connected to inferior performance of CAR T-cells.

In summary, we generated a panel of isogenic cell lines carrying mutations recurring in CLL patients. These models are indispensable for further studies of the mutations' impact on CLL therapy.

This research was partly financed by the grant MUNI/A/1224/2022 and supported by the project National Institute for Cancer Research (Programme EXCELES, ID Project No. LX22NPO5102) - Funded by the European Union – Next Generation EU, as well as by the Martina Roeselová Memorial Fellowship granted by the IOCB Tech Foundation.

P61: 3'UTR-mediated regulatory partners in MYC mRNA across different cell lines

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The 3' untranslated region (UTR) of messenger RNAs (mRNAs) is critical in regulating various mRNA-dependent processes, including post-transcriptional regulation, which controls mRNA translation, stability, and localization. The 3' UTR of mRNAs is long with secondary and tertiary structures. This makes them attractive therapeutic targets. Our understanding of the range of regulatory partners operating through the 3'UTR is limited. However, it is known that post-transcriptional regulation can be influenced by microRNA (miRNA) machinery and signaling pathway proteins, which may act in a gene-, developmental stage-, tissue-, or cell type-specific manner. The 3'UTR of MYC proto-oncogene is highly structured with several RNA motifs, making MYC an attractive therapeutic target. MYC is a potent transcriptional regulator that drives tumor development, and its dysregulation is critical in oncogenesis. MYC expression is tightly regulated, with multiple mechanisms controlling its levels. Therefore, MYC is the first target of this study. The study aims to identify the potential miRNA and RNA binding proteins (RBPs) responsible for regulating expression via its 3'UTR.

Our initial findings indicate that MYC 3'UTR significantly affects translation rates in both HeLa and HEK cells, with a more pronounced effect observed in HEK cells(Fig.1). Several miRNAs were selected based on their ability to target the MYC 3'UTR region and their higher expression rate in each cell line. Using the RNA pulldown technique in HEK-293 and HeLa cells overexpressing the MYC 3'UTR, RBPs that play a vital role in regulating and processing mRNA were enriched. Future work will extend the study to other cell lines and investigate different disease-related mRNAs to better understand mRNA regulation.

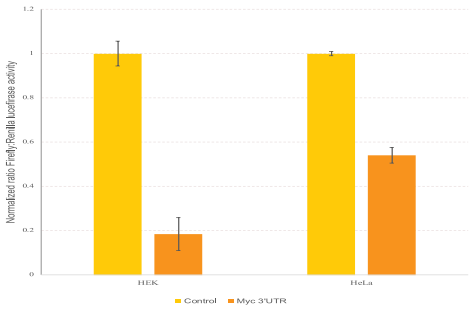

Figure1. Dual Luciferase Activity shows the impact of MYC 'UTRs on translation rates HeLa and HEK293

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P62: Unraveling the process of thermoregulation during the seed development in *Brassica napus*

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Studies on plant development under warm temperature conditions provide knowledge about the temperature's influence on crop yield. *Brassica napus* is the second most widely produced oilseed worldwide. Characterizing the thermomorphogenesis of *B. napus* grown in long-term heat stress conditions identified accelerated plant growth, reduced fertilization rate, and increased seed abortion rate. The accelerated and defective embryo development and pre-harvest seed sprouting in plants grown under heat stress suggest a possible reduction in seed dormancy. We identified a reduced expression of ABA biosynthetic genes and dormancy markers. However, the phenotypes were not reverted by external ABA applications. We hypothesized a link between high temperatures, accelerated embryo growth, and the mechano-sensing pathway during the early seed maturation phase under heat stress. Studies in this research area will pave the way toward producing thermotolerant varieties of *B. napus* with better crop yield.

P63: On the role of long non-coding RNA in BCR signaling regulation in chronic lymphocytic leukemia

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Chronic lymphocytic leukemia (CLL), the most common leukemia among adults, is largely driven by the deregulation of B-cell receptor (BCR) signaling. We aim to investigate for the first time the contribution of long non-coding RNAs (mainly LINC01480) to the deregulation of the BCR pathway. Our preliminary data suggest that LINC01480 can play an important role in tuning the activity of this pathway in CLL, which has implications for understanding disease aggressiveness and targeted therapy.

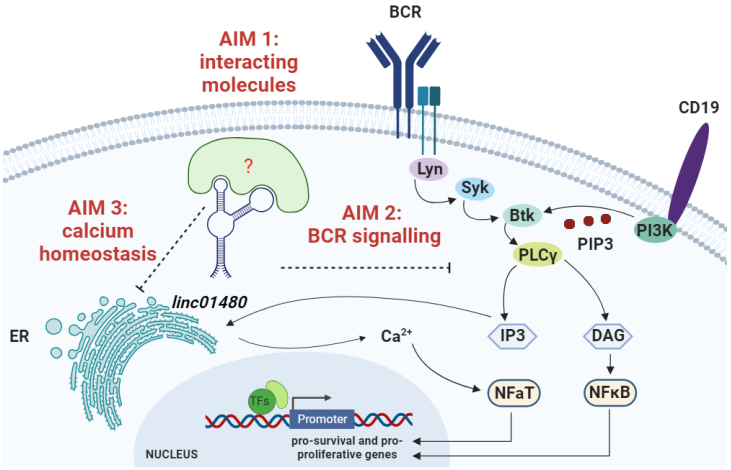

Figure: Graphical summary of the project

P64: Structural color filters with compensated angle-dependent shifts

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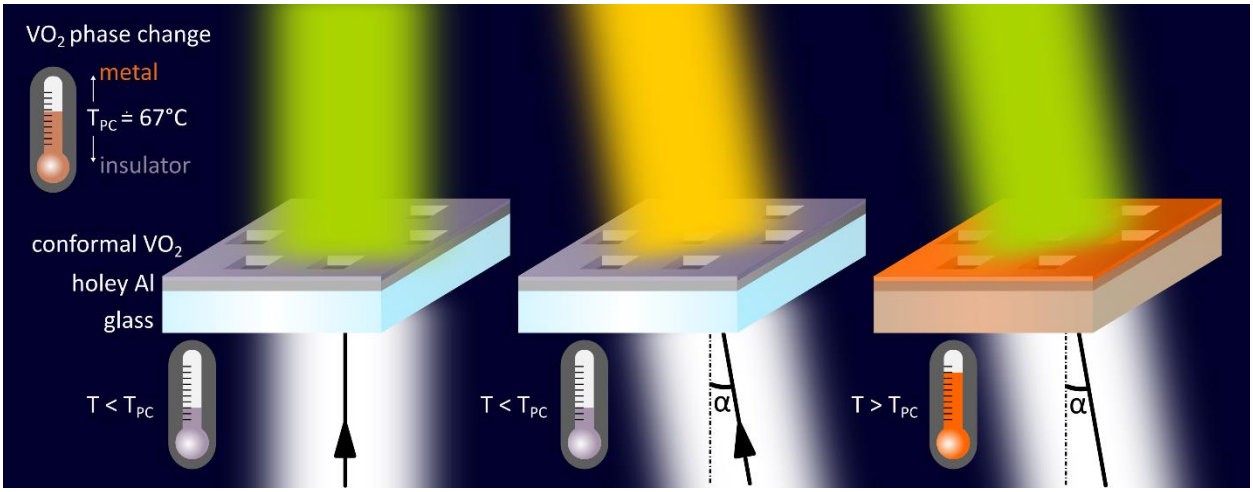
Structural color filters use nano-sized elements to allow for selective transmission of the incident light. Compared to the traditional, pigment- and dye-based color filters, they offer a cheaper, less environmentally harming and more scalable alternative of color filtering. However, their structural nature makes them very sensitive to the illumination direction and leads to spectral shifts in their optical response as the incident angle varies. So far, strategies for the mitigation of this issue relied heavily on the design of the color filter's geometry, which inevitably limited their overall efficiency, often by restricting some light polarization states. Here, we demonstrate a highly flexible compensation method for the angle-dependent spectral shifts by introducing a conformal VO_2 layer onto the bare aluminum structural color filters.
Driving the VO_2 through its insulator-metal transition has enabled us to restore the original spectral position of the color filter's transmission maximum even when the color filter was tilted from the light source by 15°, while 80% of the transmission was maintained and narrower FWHM was achieved. The key advantage of the presented compensation method lies in its flexibility: As an additional fabrication step, it can be seamlessly integrated into the manufacturing of any structural color filter, regardless of its design. Our findings establish dynamically tunable phase-change materials, such as VO_2, as a possible solution for angle-dependent spectral shifts.



P65: Plasma processing: an efficient approach for lignin particles immobilization on synthetic polymeric mats.

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Nanofibers (NFs) with their high porosity, 3D architecture and interconnected pore network have been emerged as promising scaffold materials in tissue engineering [1]. Furthermore, Nanofibers could be empowered with nanoparticles as carrier material, enabling a functional therapy by drug delivery system. Nevertheless, while synthetic materials have been widely used to overcome complexities associated with natural polymeric materials, they exhibit low surface energy. Plasmas has been extensively used method for the surface modification of polymers enabling the chemical composition for the biological attributes needed while preserving material bulk properties [2]. We have reported in previous works that plasma polymers PPs-coated surfaces have a great potential for tissue remodeling and regeneration thanks to their excellent processability, bioactivity and high reactivity allowing the formation of the covalent linkages between biomolecules and a surface [3]. Hence, we are able to introduce bio-modulated microenvironment by combining the physical architecture and the chemical composition of extra cellular matrix (ECM). This work, underscore the efficiency of plasma processing in altering synthetic polymer surface: polycaprolactone (PCL), for the immobilization of lignin nano or microparticles (fig.1). Modifications were carried out in the low-pressure capacitively coupled radio frequency discharge. We achieved consistent and densely packed attachment of lignin particles.

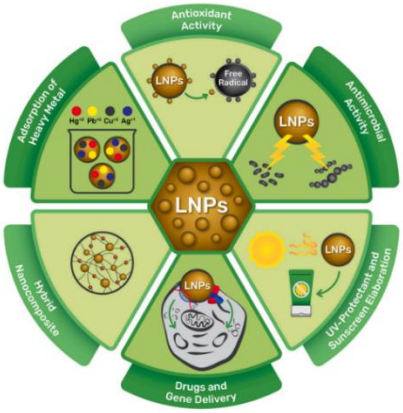

Figure 1: multi-functionality of lignin nanoparticles [4].

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P66: Mice as a genetic model to study Adar, the RNA editing enzyme

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The ADAR1 RNA-editing enzyme that deaminates adenosines (A) to inosine (I) in dsRNA is required to prevent aberrant activation of antiviral cytoplasmic dsRNA sensors by endogenous unedited dsRNA. *Adar*$^{\Delta 2\text{-}13}$ null mutant mice lacking most of the coding sequences for the Adar1 and *Adar*^{E861A} catalytically inactive mutants both die as embryos with high, aberrant interferon induction. Interferon induction is due to aberrant activation of the cytoplasmic antiviral dsRNA sensor Mda5 by unedited endogenous dsRNA. However, *Adar*^{E861A} embryos live two days longer than *Adar*$^{\Delta 2\text{-}13}$ null mutants, indicating that the retention of even an inactive Adar1 E861A offers some protection and that editing-independent functions of Adar1 is also important.

To study this editing independent functions, we compare the survival to birth, growth and lifespans of pups from parallel, genetically matched mouse strains that give different degrees of rescue of *Adar*$^{\Delta 2\text{-}13}$ null and *Adar*^{E861A} mutant phenotypes. Pups from *Adar*^{E861A}-based strains consistently show less severe defects compared to pups from matched *Adar*$^{\Delta 2\text{-}13}$. *Adar*$^{\Delta 2\text{-}13}$ *Mavs* double mutants that also lack the Mavs protein required for interferon induction downstream of cytoplasmic antiviral dsRNA sensor Mda5 avoid embryonic lethality to give live born pups as does or *Adar*$^{\Delta 2\text{-}13}$ *Ifih1*(Mda5). The *Adar*$^{\Delta 2\text{-}13}$ *Mavs* double mutants die within 10-15 days, while *Adar*$^{\Delta 2\text{-}13}$ *Ifih1*(Mda5) die sooner within 5-10 days. We also generated *Adar*^{E861A} *Ifih1* (Mda5) or *Adar*^{E861A} *Mavs* double mutant mice that appear fully rescued and reach breeding age. We are currently investigating the role of *Adar*^{E861A} in intestinal apoptosis, which occurs in *Adar*$^{\Delta 2\text{-}13}$ *Mavs* double homozygous pups but should not occur in the *Adar*^{E861A} *Mavs* double mutant. Double mutant mice with *Adar*^{E861A} and either *Mavs* or *Ifih1* mutations appear well rescued.

Aberrant activation of a second dsRNA sensor, Protein Kinase R (Pkr) can inhibit translation and drive stress granule formation, and cell death. To complete full rescue of pup survival, we generated *Adar*$^{\Delta 2\text{-}13}$ *Mavs Eif2ak2* (Pkr) triple mutants. Triple mutants grow slowly and up to 80% survive long-term and can be used for breeding. We show that the early death and severe gut defects in double mutant pups arising from death or aberrant differentiation of proliferating gut stem cells are rescued in *Adar*$^{\Delta 2\text{-}13}$ *Mavs Eif2ak2*^{Ps1} triple mutant mice

P67: Metal-organic frameworks on the topological insulator Bi_2Se_3(0001) surface

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Exotic quantum matter and in particular topological insulators have recently drawn more and more attention due to their fascinating electronic properties [1,2]. Heterostructures of these materials with other phases of matter (e.g., superconductors, organic molecules or a precise arrangement of spins/metal atoms) are highly interesting candidate structures for a variety of applications in new quantum devices and thus of considerable interest for optoelectronics, quantum computing, or spintronics [3].

In a three-dimensional topological insulator (TI) strong spin orbit coupling combined with time reversal symmetry (TRS) leads to a band gap inversion and thus to a topologically protected surface states with linear dispersion characteristic for Dirac fermions. Whereas surface perturbations that maintain TRS (e.g., surface defects or chemical impurities) cannot induce backscattering of electrons and thus do not influence the nearly dissipation less current at the surface, periodic arrays of ferromagnetically coupled transition metal (TM) atoms are predicted to spontaneously break TRS [4] and induce a band gap opening at zero magnetic field (i.e., a quantum anomalous Hall effect, QAHE). A 2D metal organic framework (MOF) of spin coupled TM atoms ordered by the right organic linkers could be such a candidate.

Here, we present an initial step to reach this goal on the topological insulator surface of Bi_2Se_3(0001). We report on the self-assembly of dicyanoanthracene (DCA) molecules on the Bi_2Se_3(0001) surface as a first step for the generation of MOFs, and first experiments of Fe-DCA MOFs on a topological insulator. We apply a variety of surface science techniques (LEEM, LEED, STM, XPS) to investigate and characterize the formation of molecular islands from small to monolayer coverages on the TI surface. This combination of techniques first allows us to study (live view) island growth at the mesoscale (LEEM) followed by information on the precise molecular arrangement at the molecular (atomic) level within one island (LEED and STM). Complementary to this, we investigate the chemical environment by in situ XPS.

Acknowledgement: This research has been funded by the European co funded IMPROVE V initiative, project No. CZ.02.01.01/00/22_010/0002552 as well as by the GAČR, project No. 22 04551S. We especially thank Čestmir Drašar from the University of Pardubice, who fabricated the used Bi2Se3 samples for us. Finally, we acknowledge the CzechNanoLab Research Infrastructure (LM2023051) supported by MEYS CR for access to the experimental facilities and financial support.

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P68: Ultrathin TiO2 ALD coatings strongly enhance biological response of biomedical materials

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TiO_2 surfaces are in general recognized as excellent biocompatible materials owing to their low cytotoxicity, high stability, antibacterial properties, and wetting ability. Among various TiO2 nanostructured surfaces that show very good cell interactions (various cell types) and osseointegration, anodized TiO_2 nanotube (TNT) layers have emerged as extremely suitable substrates. A pioneering work demonstrated that TNTs with diameter of 15 nm are the most suitable for the growth of various cells [1]. But numerous papers also showed that anodization is a very viable tool for nanostructuring of various biomedical alloys, including frequently used TiAlV.

Recently, we demonstrated that an ultrathin coating on TNT by suitable oxides (e.g. TiO2) using Atomic Layer Deposition (ALD) can enhance cell growth and adhesion [2]. These properties make them excellent as final surfaces for medical and dental implants based on Ti alloys. The presentation deals with the comparison of the influence of ultrathin ALD TiO_2 coatings (achieved by few cycles of TiO_2 ALD process) on TNT layers, reference Ti foils and Ti biomedical alloys for the proliferation of fibroblast, osteoblast and neuroblasts cells. For that Ti sheets and anodized TNT layers with a distinct inner diameter of 12 nm, 15 nm, and 100 nm were used as substrates, as they appear to be the most suitable for cell growth in general [2,3,4]. We investigated the shaping, adhesion, proliferation, and cell density on these substrates. Moreover, the single-cell adhesion of the cells to the TNTs was studied by the Bio-atomic force microscopy (bio-AMF) technique [3]. Last, but not least, black form of TNTs was investigated for cell proliferation in comparison to classical TNTs [4].

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P69: A facile one-pot synthesis of water-soluble CQDs for the evaluation of their anti-amyloidogenic propensity

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In this study, we present a straightforward microwave-assisted one-pot synthesis method to create highly water-soluble CQDs. We investigate their ability to hinder amyloid fibrillation and their potential as a drug for addressing protein-related neurodegenerative disorders. These CQDs, with an average size of 7 nm, are synthesized by directly carbonizing folic acid and bis-acrylamide under microwave conditions. They exhibit a prolonged shelf life due to their substantial negative surface charge, enabling exceptional water solubility. This characteristic leads to high photo-stability, a high quantum yield, all achieved without the need for post-synthesis surface passivation steps, simplifying the process compared to contemporary methods. Upon examination, the synthesized CQDs exhibit significant inhibition of amyloid aggregation in hen egg-white lysozyme (HEWL) under both acidic and neutral pH conditions. They can inhibit up to 60% of the aggregation process in acidic pH and up to 50% in neutral pH, as confirmed by various assays, including ThT and ANS. Under certain destabilizing conditions, proteins tend to unfold and form ordered β-sheeted fibrillar aggregates known as "amyloids." These amyloid fibrils are critical precursors in protein-linked degenerative diseases such as Alzheimer's disease, Parkinson's disease, Huntington's disease, and Type II diabetes mellitus. Given the anti-amyloidogenic properties of these CQDs and their outstanding intrinsic characteristics, they hold promise as potential candidates for the treatment of protein-related neurodegenerative diseases.

P70: Electron Tractor Beam: Deterministic Manipulation of Liquid Droplets on Solid Surfaces

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Motion of liquid droplets is studied across many subfields of physics and manipulation of nanoscale and microscale droplets on demand holds great promise in e.g., nanotechnology[1,2]. In this study, AuGe droplets on germanium substrate are manipulated by an electron beam in scanning electron microscope. The electron beam exposure creates a local temperature gradient in a substrate and induces a directional thermomigration of droplets nearby. To quantitatively analyze this phenomenon and to reveal the mechanism behind, experimental observations under different conditions (beam current, sample temperature, scanning strategy) and simulations were combined. The obtained insights suggest that the droplet motion is limited by the dissolution of the substrate below the droplet. In addition, it was shown that the electron tractor beam is not only able to control the droplet motion, but can also be used to split the droplets, thus opening new possibilities for droplet manipulation inside a scanning electron microscope (SEM).

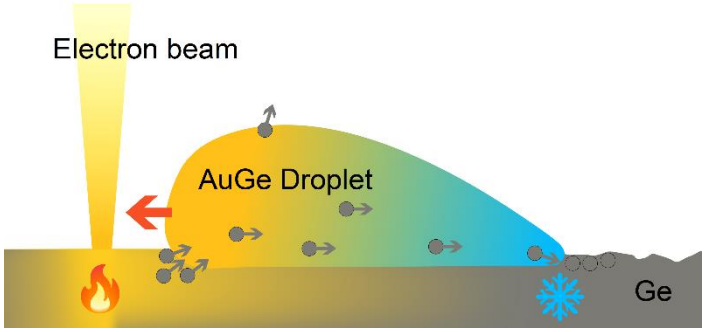

Figure 1. AuGe droplet on Ge substrate with temperature gradient, which is caused by heating from electron beam. On the warmer side of the droplet, Ge solubility is increased, thus Ge atoms (gray circles) are absorbed from substrate into the droplet. Inside the droplet, the Ge atoms diffuse according to Fick's law, and they are deposited on the colder side of the droplet. In consequence, the droplet is moving toward the warmer area, i.e., the droplet is following the electron beam.

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P71: Self-Assembly of Bile Acid Derivatives into Metallosupramolecur Architectures

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Natural chiral hydrophobic cavity/pocket containing structures (e.g., metalloenzymes, proteins) are important for many biological functions (e.g., transport, recognition, catalysis). To mimic these natural systems and mechanisms, development of such supramolecular systems (e.g., cages, macrocycles) from chiral natural molecules is required.

Coordination-driven self-assembly is a well-established method to build hollow metallosupramolecular (MSM) structures. However, majority of self-assemblies are made of symmetric, achiral ligands (L) and Pd^{2+}. Recently in our group, first bile acid (BA)-based (ursodeoxycholic acid, UDCA) MSM macrocycles Pd_3L_6 (Figure 1a) were introduced[1] and studied.[2] Beside this, there is only one report about BA-based Pd2L4 MSM cages.[3] We further expanded on the family of BAs by synthesizing chenodeoxycholic acid-based ditopic pyridyl ligand, forms a mixture of Pd_nL_{2n} species ranging from Pd_2L_4 to a large Pd_6L_{12}.

Thus far, only BA-based ditopic pyridyl ligands were used to prepare MSM systems. Therefore, our latest study presents UDCA-based tritopic pyridyl ligand and its self-assembly with Pd^{2+}, which results in Pd_6L_8 (Figure 1b) or first-ever giant $Pd_{12}L_{16}$ MSM cage depending on solvent and metal-ligand ratio.

These studies provide better understanding of unsymmetric natural molecule-based ligands self-assembly, effect of their flexibility, topicity, and bend angle in design and construction of chiral cavity containing MSM architectures.

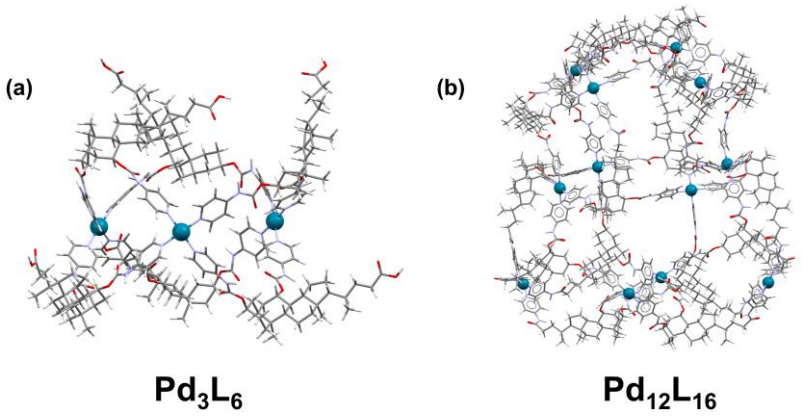

Figure 1. Bile acid-based metallosupramolecular a) macrocycle and b) cage.

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P72: Methyl Germanane Enhanced 3D-Printed Nanocarbon Electrode as a Photoelectrocatalyst for Hydrogen Evolution Reaction

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Two-dimensional (2D) layered materials beyond graphene are currently the focus of extensive investigations. After the massive exploration of graphene in widespread applications, other elements of group 14, including germanene became the topic of interest as potential monoelemental Xene material. Interestingly, 2D layered germanene can be modified with covalent functionalization, including hydrogen- or methyl- or other alkyl-groups termination, to obtain germanane, which offers tunable physical, chemical, optical, or electronic properties. The stability and the photoactivity in the visible spectral region of methyl germanene (Ge-CH_3) are promising characteristics for optoelectronics and photoelectrochemical applications[1].

Three-dimensional (3D) printing is a widespread method to print 3D objects with free choice of size and shape by adding the material layer-by-layer. Among many additive manufacturing techniques, low cost, low wastage of material, and easiness of usage make fused deposition modeling (FDM) one of the best 3D printing options. 3D conductive carbon structures are widely prepared by extruding carbon-thermoplastic filament[2]. However, as printed 3D nanocarbon structures usually do not have required properties and typically need to be modified with active materials to introduce such properties. In this work, we show how 2D methyl germanane enhances 3D printed carbon electrodes as photoelectrocatalyst for hydrogen evolution reaction (HER).

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P73: Electrochemical processes at clinical PtIr stimulation electrodes

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One of the main purposes of neural bioelectronic devices is to deliver electrical stimulation, which is typically conducted applying biphasic pulses: the first pulse is cathodic and has the aim to depolarize the cell inducing an action potential, the subsequent anodic pulse should convert the unintended redox products to their original form. However, such electrochemical processes are never fully reversible, in particular, oxygen reaction reduction (ORR) and hydrogen evolution reaction (HER), can induce oxidative stress and affect the pH.
Reactive oxygen species (ROS), oxygen concentration and acid-base homeostasis are regulated in all living systems, this is critical because even a slight change in pH or oxidative stress can heavily affect cell physiology and functions, as well as leading to cell death. Although it is well known that living systems have fast response to such changes, the effect of localized reactions is still poorly explored.
The aim of this work is to quantify O_2 and H_2 concentration in proximity of the electrode and the related change in pH when electrical neurostimulation protocols are applied. PtIr stereo electro encephalography (SEEG) electrodes were chosen because they represent electrode material used in the majority of clinically approved implantable electrical stimulators. In order to map pH changes, a potentiometric microsensor was used to scan the pH at different distances from the SEEG electrode while a wide range of potential (vs. Ag/AgCl) are applied. Moreover, to compare the effect in different media, the experiment was conducted in a standard buffered solution (PBS) in an inert unbuffered solution (Na_2SO_4) and in cell medium.

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P74: Study of the involvement of the PHYB-PIF4 pathway in high-temperature responses during the reproductive phase and embryogenesis of *Arabidopsis thaliana*

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The global climate system is warming up, and the increase in temperatures is one of the important factors affecting multiple developmental processes, like growth and flowering. Using Arabidopsis thaliana as a model organism substantially increases our knowledge of the signaling and response mechanisms during plant exposure to elevated temperatures. A substantial amount of data has been gathered to elucidate the role of hormones and molecular chaperones, such as Heat shock proteins (HSPs), in this temperature response in vegetative tissues. In this study, we analyze the function of the PHYB-PIF4 pathway, known for controlling high-temperature stress in the seedling during reproductive development and seed production. For this purpose, some mutants [phyb, pif4, and quadruple pif1/3/4/5 mutants (pifq)] and a PIF4 over-expression line have been studied. Embryo phenotyping demonstrates that heat stress causes suspensor shortening. Further analysis is required to elucidate if PIF4 is involved in this response. Besides dwarf suspensor, we observed some defects in the embryo proper division at high temperatures, such as extra division in the hypophysis. The severity of the heat stress effect was roughly the same in all the studied lines. Although reanalysis of RNA-seq data and histochemical GUS assays showed that PIF4 is not expressed during embryonic developmental stages, we show that elevated temperature increases PIF4 expression in the seed integuments. Microscopy analysis of YUCCAs reporter lines showed that temperature also alters the expression level of some auxin biosynthesis genes (YUCCA4, YUCCA8) in the seed coat. We hypothesize that higher PIF4 expression in the integuments enhances the YUC8 expression causing abnormal embryo divisions. Ovule phenotyping indicates that over-expression of PIF4 at normal conditions mimics the effect of heat stress on the wild type. Furthermore, exposure of the phyb mutant and p35S:PIF4 lines to heat stress results in defects of around 90 % of produced ovules, while this percentage decreases to about 40 in wild-type, pif4, and pifq mutant lines. GUS assays indicate that elevated temperatures boost PIF4 expression in the ovule too. We will perform further experiments like CUT&RUN-seq to find PIF4 target genes at high temperatures in ovules and seeds.

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P75: The Role of Components of Cytokinin Metabolism in *in vitro* Shoot Regeneration

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Plants possess unique developmental plasticity, which results in their efficient regeneration capacity, including *in vitro* regeneration of plant organs. Such processes are strictly regulated by the activity of phytohormones and the fine-tuned cooperation of some groups of phytohormones are often necessary. During shoot regeneration the presence of two phytohormone groups, auxin and cytokinins, is detrimental. When explants are placed on the medium containing auxin, the formation of primordium of the root identity is triggered. When a presence of auxin is prevalent, the roots are developed. In case of cytokinin presence, the root primordium undergoes developmental switch and regenerates into shoot[1]. The participation of cytokinins in this developmental switch is essential. In our work, the attention was aimed at components of the cytokinin biosynthetic pathway. Particularly, we focused on LONELY GUY (LOG) family members - enzymes responsible for the last critical step in biosynthesis of active cytokinins[2,3].

We analyzed shoot morphology of *log* mutant lines using phenotyping. The results showed that mutants in *LOG4* gene which is expressed in L1 layer of the shoot apical meristem and in *LOG3* gene, which is active in developing leaf primordia[4], showed altered shoot morphology. During the process of *in vitro* shoot regeneration, the shoots first regenerate directly via the activity of the pericycle cells. The most of newly regenerated shoots were already recognizable after 9 days of regeneration. In the meantime, the callus was formed on the explants out of which indirectly regenerated shoots emerge after 15 days of regeneration. However, *in vitro* shoot regeneration assay did not reveal any visible differences of *log* single and multiple mutants in their ability to regenerate shoots. This refers to their functional redundancy or possible involvement of another cytokinin biosynthetic pathway.

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	Name	Surname	Poster/talk no	page
1	Mahnaz	Alijani	P1	30
2	Damiano	Amorouso	P42	72
3	David	Antoš	P28	58
4	Mahya	Asharian Marzabad	P36	66
5	Kaushik	Baishya	P68	98
6	Nilanjan	Banerjee	P2	31
7	Samia	Belaidi	P40	70
8	Inderjeet	Bhogal	T12	23
9	Denys	Biriukov	T1	11
10	Hatef	Bokaei Khelejan	P49	79
11	Alicia	Burkard	P47	77
12	Eva	Černa	T17	28
13	Subhasis	Chattopadhyay	P71	101
14	Anna	Cherian	T9	20
15	Valli Kamala Laxmi Ramya	Chittoory	T10	21
16	Lenka	Dostálova	P37	67
17	Lenka	Drotárová	P48	78
18	Qiupei	Du	P56	86
19	Jan	Dubský	P29	59
20	Zuzana	Dvořáková	P3	32
21	Shekoufeh	Ebrahimi Naghani	P74	104
22	Jiří	Ehlich	P4	33
23	Petr	Fajkus	T2	12
24	Jan	Farkaš		
25	Sanam	Garehbaghi	T14	25
26	Khadija	Hajji	P5	34
27	Samer	Halabi	T3, T5	13, 15
28	Klára	Hánělová	P30	60
29	Alena	Hofrová	P43	73
30	Jakub	Holobrádek	T6	17
31	Beáta	Idesová	P24	54
32	Mohd	Isar	P59	89
33	Dávid	Izsák	P51	81
34	Marie	Jakešová	T4	14
35	Pawel	Jewula	P6	35
36	Jana	Juráková	P31	61
37	Péter	Kacz	P63	93
38	Zuzana	Kadlecová	P32	62
39	Adelia	Kashimbetova	P41	71
40	Lea-Marie	Kilz	P52	82
41	Hana	Kopřivová	P23	53
42	Helene	Kristiansen	P27	57
43	Anna	Kurowská	P67	97
44	Valentina	Lacovich Strašil	P7	36
45	Oleksii	Laguta	P8	37
46	Adela	Lamaczova	P33	63
47	Jarmila	Laušová	P34	64
48	Kateřina	Linhartová	P35	65

49	Sepideh	M.Koubjari	P61	91
50	Martin	Marônek	P9	38
51	Michaela	Medková	P22	52
52	Janka	Melicherová	P66	96
53	Ivana	Michálková	P25	55
54	Tereza	Mikešová	P53	83
55	Noor	Mohamad	P54	84
56	William	Morton	P10	39
57	Aniket	Mukherjee	P69	99
58	Hana	Nedozrálová	P11	40
59	Lucie	Nepovímová	P39	69
60	Radhika	Nittoor Veedu	P72	102
61	Katarina	Novčić	P44	74
62	Ales	Obrdlik	P12	41
63	Petra	Ondráčková	P13	42
64	Vaishali	Pankaj	T8	19
65	Peter	Patjinka	T16	27
66	Shib Shankar	Paul	P46	76
67	Pavel	Payne	P14	43
68	Helena	Peschelova	P60	90
69	Michaela	Pešová	T7	18
70	Veronika	Poláková	P55	85
71	Unnikannan	Prabhullachandran	P62	92
72	Alena	Raszková	P57	87
73	Katarína	Rovenská	P64	94
74	Amedeo	Ruggiero	P73	103
75	Michaela	Růžičková	T11	22
76	Arezoo	Saadati	P26	56
77	Surendra	Saddala	T13	24
78	Mateo	Seoane Blanco	P15	44
79	Azin	Shahsavar	P16	45
80	Neha	Shukla	P17	46
81	Nada	Souawda	P65	95
82	Iva	Sukkar	P18	47
83	Ján	Šmeringai	P75	105
84	Denis	Šubert	T15	26
85	Muhammad	Tahsin	P38	68
86	Vlastimil	Tichý	P19	48
87	Anna	Tvrdoňová	P58	88
88	Iveta	Ukropcová	P70	100
89	Mohammad	Umair		
90	Marlies	Weber	P45	75
91	Ikram	Zdeg	P50	80
92	Marichka	Zlatohurska	P20	49
93	Maria	Zlobina	P21	50

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