LIÈGE université
GIGA institute

2024 GIGA DAY

13 SEPT
FRIDAY

AMPHIS
EUROPE
B4 SART-TILMAN

ABSTRACT BOOK

INVITED SPEAKERS

MASSIMILIANO MAZZONE, VIB, KU LEUVEN
GERALDINE PIEL, CIRM, ULIÈGE
VEERLE ROTS, TRACEOLAB, ULIÈGE
VINCENT PREVOT, INSERM LILLE
JUDITH COSEMANS, MAASTRICHT UNIVERSITY

GIGA DAY
13 SEPT

PROGRAM

08:30 Welcome coffee

09:00-09:40 **Massimiliano Mazzone, VIB, KU Leuven**
Harnessing metabolism to increase the success of cancer immunotherapies

09:40-10:00 **Raphaël Peiffer, Cancer**
Myoferlin participates in TGFß-receptor trafficking in cancer-associated fibroblasts, promoting tumor desmoplasia and stromal aggressiveness in pancreatic cancer

10:00-10:20 **Cécilia Ruscitti, Immunobiology**
Recruited atypical Ly6G+ macrophages license alveolar regeneration after lung injury

10:20-11:00 Coffee Break

11:00-11:40 **Geraldine Piel, CIRM, ULiège**
Liposomes and lipid nanoparticles as non-viral vectors for drug delivery: advantages, limitations and challenges

11:40-12:00 **Marjorie Lienard, Molecular & Computational Biology**
Molecular mechanisms of spectral tuning in Gq-coupled rhodopsins

12:00-12:10 Flash talks

12:10-13:30 Lunch Break

13:30-14:10 **Veerle Rots, TraceoLab, ULiège**
Tracing Neanderthals and early modern humans through the microscope

14:10-14:30 **Carmen Cabello, Neuroscience**
Existence of different profiles in long covid patients with cognitive complaints

14:30-15:10 **Vincent Prevot, Inserm Lille**
Pulsatile secretion of GnRH and cognition: a role for minipuberty

15:10-15:40 Coffe break

15:40-16:20 **Judith Cosemans, Maastricht University**
Exploring platelet and coagulation function in heart failure with preserved ejection fraction

16:20-16:40 **Sofia Melo, Metabolism & Cardiovascular Biology**
Sustainable polymers with improved biocompatibility: new materials for blood-contacting medical devices

16:40-17:00 Poster prizes / end

Invited Speakers

Massimiliano Mazzone
Center for Cancer Biology
KU Leuven

Massimiliano (Max) Mazzone graduated in Medical Biotechnology at the Medical School of the University of Torino, Italy. Since he started his independent research at VIB, he focused on the response of inflammatory cells to hypoxia, metabolites and cytokine surges, aiming to restore blood flow in conditions as cancer and ischemia. He pioneered the concept that localization of tumor-associated macrophages (TAMs) is determinant for their pro-vascular function, immune phenotype and for the anti-tumor T cell response and has shown that some of these inflammatory fingerprints can be exploited for disease detection and follow-up in cancer patients or can be used to predict disease response to targeted therapies. As a result of his research, qualitative rewiring of innate immunity has replaced the original concept of quantitative inflammatory cell disruption. With his new concept that hypoxic TAMs are the pro-tumoral TAMs that have to be turned off while feeding the anti-tumor properties of TAMs in normoxia, he moved away from the more dogmatic definitions of M1 and M2 polarized macrophages. Indeed, his work shows that hypoxia fine-tunes the immune phenotype of macrophages without affecting the expression of canonical polarization markers. As well, a subset of TAMs characterized by surface markers and a specific metabolism cuffs the lymphatic vessels in the tumors promoting new sprouts and favouring lymphoinvasion and lymphatic metastasis.

HARNESSING METABOLISM TO INCREASE THE SUCCESS OF CANCER IMMUNOTHERAPIES

Anti-cancer immunotherapy has provided patients with a promising treatment. Yet, it has also unveiled that the immunosuppressive tumor microenvironment (TME) hampers the efficiency of this therapeutic option and limits its success. The concept that metabolism is able to shape the immune response has gained general acceptance. Nonetheless, little is known on how the metabolic crosstalk between different tumor compartments contributes to the harsh TME and ultimately impairs T cell fitness within the tumor. This lecture will decipher some of the metabolic changes in the TME impeding proper anti-tumor immunity. Starting from the meta-analysis of public human datasets, corroborated by metabolomics and transcriptomics data from several mouse tumors, we ranked clinically relevant and altered metabolic pathways that correlate with resistance to immunotherapy. Using a CRISPR/Cas9 platform for their functional in vivo selection, we have identified cancer cell intrinsic metabolic mediators and, indirectly, distinguished those belonging specifically to the stroma. By means of genetic tools and small molecules, we have targeted promising metabolic pathways in cancer cells and stromal cells (particularly in tumor-associated macrophages) to harness tumor immunosuppression. Finally, we went back to patient samples to assess the relevance of these metabolic networks in humans. By analyzing the metabolic crosstalk within the TME, this lecture would like to shed some light on how metabolism contributes to the immunosuppressive TME and T cell maladaptation.

Géraldine Piel
CIRM
ULiège

Geraldine Piel is a pharmacist (1993) and holds a PhD in Pharmaceutical Sciences from the University of Liège (2000). After her PhD thesis, she continued her training with a post-doctoral stay at the University of Paris-Sud in the laboratory of Professor Elias Fattal on the vectorization of fragile molecules, where she acquired extensive knowledge of nanomedicine development techniques. Upon her return to Liège, she initiated and developed various aspects of nanomedicine formulation, with a particular focus on the development of more effective and patient-friendly therapies based on lipid-based nanoparticles.
Her research includes significant contributions to the development and characterization of lipid nanoparticles, the use of supercritical CO2 for liposome production and sterilization, and the formulation of drugs for the treatment of specific conditions such as sensorineural hearing loss and pulmonary diseases.Her research has resulted in more than 100 publications and numerous presentations at various scientific forums and conferences.

LIPOSOMES AND LIPID NANOPARTICLES AS NON-VIRAL VECTORS FOR DRUG DELIVERY: ADVANTAGES, LIMITATIONS AND CHALLENGES

This talk proposes to give an overview of the advantages, limitations and challenges of lipid nanoparticles, also called non-viral vectors, for the delivery of different types of active molecules, from small classical pharmaceutical molecules to genetic materials, for different routes of administration (both systemic and topical). The presentation will be illustrated with examples of developments that have been made or are in progress in the Pharmaceutical Technology Laboratory.

Veerle Rots
TraceoLab
ULiège

Veerle Rots obtained her PhD at KULeuven and subsequently became a postdoctoral research fellow of the Research Fund (2002-2009) and the Fund for Scientific Research of Flanders (FWO) (2009-2011). She was appointed guest lecturer at the Department of Archaeology of the University of Leuven in 2005 until 2012. In 2011, she obtained a permanent position as researcher of the Belgian Fund for Scientific Research (FNRS-FRS) at the University of Liège (Belgium). She was awarded a prestigious ERC starting grant form the European Research Council in 2012 with the project «Evolution of Stone Tool Hafting in the Palaeolithic». In 2019, she became a Senior Research Associate of the F.R.S.-FNRS. She was awarded the Francqui prize in 2022. Currently, she is Research Director of the F.R.S.-FNRS (since 2023). Since her arrival at the University of Liège, she has developed TraceoLab, a research centre in prehistory devoted to use-wear and residue studies of prehistoric stone tools and experimentation. Her personal expertise mainly concerns integrated techno-functional approaches with a particular focus on microscopic use-wear and hafting wear traces on stone tools, associated with systematic experimentation, and with a particular interest in Palaeolithic assemblages. Over the years, she has been involved in many field projects in Belgium and abroad (e.g., Egypt, Sudan, Ethiopia, Turkey, Poland, South Africa), and she has on-going collaborations to examine the archaeological material of different Palaeolithic sites (Belgium, Germany, France, Italy, Morocco, Ethiopia, South Africa, Zambia).

TRACING NEANDERTHALS AND EARLY MODERN HUMANS THROUGH THE MICROSCOPE

About 3.3 million years ago, humans started to produce the first stone tools and they have continued to invest in their technology ever since. Since organic components of prehistoric technology rarely preserve, stone tools are the most common finds at prehistoric sites. Stone tools and the microscopic evidence they hold therefore represent a crucial source of information to understand prehistoric lifeways and how human behaviour evolved over time. Indeed, a microscopic investigation of stone tools enables us to discover how tools were used, what the function of prehistoric sites was and how hunter-gatherer groups were organised across a landscape. Such a functional approach also permits access to understanding the technology of Neanderthals and early modern humans and evaluating to what extent their behaviours were distinct. Do they share a capacity for long-term planning and the manufacture of complex technologies? Did they both fit stone tools into handles and why would this be important? Did they use similar hunting weapons, how could we know, and what would be the implications? This presentation takes you on a quest across several continents to find out more about the behaviour of prehistoric humans, based on recent knowledge gained through the detailed microscopic examination of stone tools.

Vincent Prevot
Lille Neuroscience & Cognition
Inserm

Vincent Prevot is currently Senior Research Director at the Inserm (the French National Institute of Health and Medical Research) and Head of the «Development and Plasticity of the Postnatal Brain» laboratory at the Lille Neuroscience & Cognition Research Center in Lille, France, since 2007. The two principal focuses of his research are the Central Control of Energy Homeostasis and the Neurobiology of Reproduction. Among his recent pioneering studies is the demonstration that tanycytes, specialized ependymoglial cells lining the floor of the third ventricle, transport circulating metabolic signals such as leptin, as well as anti-obesity drugs like liraglutide, across brain barriers into the hypothalamus to regulate energy homeostasis.

AGE-RELATED LOSS OF GNRH EXPRESSION AND RHYTHMIC RELEASE IN COGNITIVE DISORDERS: A ROLE FOR MINIPUBERTY?

Pulsatile secretion of gonadotropin-releasing hormone (GnRH) is essential for activating and maintaining the function of the hypothalamic-pituitary-gonadal (HPG) axis, which controls the onset of puberty and fertility. Two provocative recent studies suggest that, in addition to controlling reproduction, the neurons in the brain that produce GnRH are also involved in the control of postnatal brain maturation, odor discrimination, and adult cognition. I will discuss the development and establishment of the GnRH system, and especially the importance of its first postnatal activation, a phenomenon known as minipuberty, to its later functions, reproductive and non-reproductive. In addition, I will discuss the beneficial effects of restoring physiological, i.e. pulsatile, GnRH levels on olfactory and cognitive alterations in Down syndrome and preclinical models of Alzheimer's disease, as well as the risks associated with long-term continuous, i.e. non-physiological, GnRH administration in certain disorders. Finally, I'll discuss the intriguing possibility that pulsatile GnRH therapy may hold therapeutic potential for the management of some neurodevelopmental cognitive disorders as well as pathological aging in the elderly.

Judith Cosemans
School for Cardiovascular Diseases
Maastricht University

Judith Cosemans is Associate Professor and Head of the Platelet Research Group at Maastricht University. Dr. Cosemans developed an interest in platelet biology during her PhD, which focused on their role in macrovascular thrombosis. Subsequently, she pursued an award-winning postdoc project, where she worked on developing flow chamber technology as an alternative to animal thrombosis models. Afterward, she received personal grants from the Dutch Heart Foundation (DHF) and a Vidi grant, enabling her to establish an independent research line on the interface of platelets and vascular biology. In 2023, she was awarded the BHF-DZHK-DHF International Cardiovascular Research Partnership Award, supporting her current research into the role of platelets in microvascular dysfunction and novel therapeutic interventions.

EXPLORING PLATELET AND COAGULATION FUNCTION IN HEART FAILURE WITH PRESERVED EJECTION FRACTION

Heart failure with preserved ejection fraction (HFpEF) results from a complex interplay of systemic syndromes, including diabetes and hypertension, characterized by chronic low-grade inflammation and microvascular dysfunction (MVD). In recent years, the importance of platelets in vascular inflammation and endothelial dysfunction emerged, suggesting platelets' involvement in MVD. However, the role of platelets in HFpEF is still poorly examined. In this presentation, data from a study investigating the potential role of platelets and the coagulation system in HFpEF will be displayed and discussed.

GIGA Speakers

Abstracts

Raphaël Peiffer

Cancer

Metastases Research laboratory

Myoferlin participates in TGFß-receptor trafficking in cancer-associated fibroblasts, promoting tumor desmoplasia and stromal aggressiveness in pancreatic cancer

Intracellular vesicle trafficking is an evolutionary conserved process implicated in a great variety of cellular functions and diseases. Pancreatic cancer (PAAD) cells exploit vesicle trafficking via the Golgi apparatus to support cellular flexibility and tumor aggressiveness by relying on vesicle-trafficking proteins such as myoferlin. However, our understanding of myoferlin-dependent vesicle trafficking is limited to cancer cells, while the function of myoferlin in the pancreatic tumor microenvironment, notably cancer-associated fibroblasts (CAFs), has been overlooked. Here we combine PAAD whole-tumor and single-cell transcriptomic analyses with immunohistochemistry to link stromal myoferlin to tumor aggressiveness. Using 2D and 3D in vitro models of human CAFs, we identify CAF-specific functions of myoferlin, as MYOF-depleted (MYOFKD) CAFs present impaired activity and reduced extracellular matrix (ECM) production. Analysis of intracellular vesicles in MYOFKD CAFs identifies myoferlin as novel functional member of COP2-coated vesicle trafficking between the endoplasmic reticulum (ER) and Golgi apparatus. Accordingly, MYOFKD causes a TGFß-receptor 1 (TGFBR1) trafficking blockade at the ER/Golgi interface, leading to altered TGFBR1 activation, impaired signal transduction, loss of ECM production and reduced stroma aggressiveness. Orthotopic transplantation of MYOFKD CAFs in mice impairs tumor establishment, while pharmacological targeting of myoferlin reduces tumor desmoplasia in tumor-bearing mice. Overall, we propose TGFBR1 trafficking as novel target to reprogram CAFs, tackle desmoplasia and control stromal aggressiveness in PAAD.

Cécilia Ruscitti

Immunobiology

Laboratory of Immunophysiology

Recruited atypical Ly6G+ macrophages license alveolar regeneration after lung injury

The lung is constantly exposed to airborne pathogens and particles that can cause alveolar damage. Hence, appropriate repair responses are essential for gas exchange and life. Here, we deciphered the spatiotemporal trajectory and function of an atypical population of macrophages after lung injury. Post–influenza A virus (IAV) infection, short-lived monocyte-derived Ly6G-expressing macrophages (Ly6G+ Macs) were recruited to the alveoli of lung perilesional areas. Ly6G+ Macs engulfed immune cells, exhibited a high metabolic potential, and clustered with alveolar type 2 epithelial cells (AT2s) in zones of active epithelial regeneration. Ly6G+ Macs were partially dependent on granulocyte-macrophage colony-stimulating factor and interleukin-4 receptor signaling and were essential for AT2-dependent alveolar regeneration. Similar macrophages were recruited in other models of injury and in the airspaces of lungs from patients with suspected pneumonia. This study identifies perilesional alveolar Ly6G+ Macs as a spatially restricted, short-lived macrophage subset promoting epithelial regeneration postinjury, thus representing an attractive therapeutic target for treating lung damage.

Marjorie Liénard

Molecular and Computation Biology

Laboratory of Molecular Biology and Functional Evolution of Sensory System

Molecular mechanisms of spectral tuning in Gq-coupled rhodopsins

The ability to capture incoming wavelengths of light and perceive the colours of the world stems from peripheral light-sensitive G-protein coupled opsin receptors (GPCRs). Using molecular and functional approaches including a robust cell-based opsin expression platform, transcriptomics, modelling and structural mutagenesis, we explore how evolution has favoured astounding solutions to gather light in (in)visible parts of the spectrum beyond human vision, across a range of invertebrate systems – from deserts to the tropics, from shallow ponds to deep seas. By populating a database of experimental opsin genotype-phenotype relationships, including mapping and assaying the effect of new mutations absent in Gt vertebrate opsins, our results contribute to uncover key functional novelty, derive general molecular mechanisms and generate the necessary data to train models focused on AI predictions, machine learning, or improving protein stability. The systematic exploration of molecular-evolutionary patterns governing phenotypes remains foundational for fields as diverse as de-novo light-sensitive protein bioengineering and biomedical optogenetics.

Carmen Cabello

Neurosciences

Laboratory of Aging & Memory

Preliminary results of COVOCG study: Profiles of Long COVID patients and effects of two psychoeducation interventions

Following a SARS-CoV-2 infection, a large number of patients experience cognitive difficulties, both objectively measured (1) and subjectively reported (2), even 24 months after the acute phase of Covid-19. Factors contributing to these cognitive difficulties are probably multifaceted including neurobiological, somatic and psychological causes (3). In addition, different profiles of Long COVID patients had been identified based on their cognitive performances (4). Accordingly, origin and trajectory of the difficulties as well as the type of intervention most beneficial for each of them, may differ. In the COVCOG study, we aim to characterise Long COVID patients based on subjective and objective measures of cognitive functioning, and to determine the cognitive intervention programmes best suited to this population (psychoeducation focusing on cognitive or psychological aspects). Data from 122 patients (aged 46.9 ±10.1 [range 21-66]; 30.3% males; time since infection 21 months ±8.6 [range 4-39]) with cognitive complaints following one or more SARS-CoV-2 infections were analysed. Participants were randomly assigned to one of the intervention groups (i.e. cognitive or affective psychoeducation) with 4 sessions of 1h30 and a reactivation session of 30 minutes a month after. Before the intervention, as well as 2 months and 8 months post-intervention, a comprehensive neuropsychological evaluation was carried out covering attentional, memory, language and executive processes, as well as self-reported questionnaires addressing cognitive complaints, fatigue, sleep difficulties, quality of life, psychological distress and impact on daily activities. Exploratory Latent Profile Analysis (LPA) and robust ANOVAs were conducted. LPA revealed three distinct profiles of patients and an improvement was observed at 2 months post-intervention for both interventions. Discussion will focus on the different patient profiles and the preliminary results at 2 months post-intervention.

(1) Tavares-Júnior et al. 2022 Cortex
(2) Han et al., 2022 Pathogens
(3) Diar Bakerly et al. 2024 Int. J. Environ. Res. Public Health
(4) Voruz et al., 2022 Brain communications

Sofia Melo

Metabolism & Cardiovascular Biology

Laboratory of Cardiology

Sustainable polymers with improved biocompatibility: new materials for blood-contacting medical devices

One of the main goals of the Laboratory of Cardiology at GIGA is to develop new polymeric heart valve (HV) prostheses, a need that has been identified over the past few years. Indeed, the effective treatment for most patients with HV disease is valve replacement by implantation of mechanical or biological prostheses. However, mechanical valves represent high risk of thromboembolism, and biological prostheses are prone to early degeneration. The emergence of a non-thrombogenic, durable, polymeric HV therefore appears as a promising solution. Polyurethanes (PUs) have adjustable mechanical properties which make them suitable for a wide range of applications, including in the biomedical field. Historically, these PUs have been synthesized from toxic isocyanates, representing a health threat. This has encouraged the search for safer and more environmental-friendly synthetic routes, leading today to the production of non-isocyanate polyurethanes (NIPUs).
In a first approach, thermally crosslinked NIPU networks were synthesized, tested in vitro, and used to produce aortic HVs. Injection molded NIPU valves were produced and tested in a pulse duplicator, showing compliant hydrodynamic performances, comparable to bioprostheses used in clinics. Subsequently to this work, other NIPUs, more adequate for easily adaptable manufacturing techniques (e.g. 3D printing), were explored. Given the unique viscous properties of some of these NIPUs, formulations for light-based 3D printing were successfully developed and objects were printed with a resolution down to the micrometer scale. Finally, a more recent class of NIPUs was synthesized, aiming to use heat-based additive manufacturing to process such biomaterials. A new thermoplastic elastomer was produced, processed by different manufacturing techniques, and evaluated in vivo after subcutaneous implantation in rabbits. All the developed NIPUs showed remarkable hemo/biocompatibility in vitro, key for blood-contacting applications.

Posters

Abstracts

Poster 1

Pierre Adam

pierre.adam@uliege.be

Laboratory of Translational Gastroenterology
Immunobiology

Characterization of the impact of NKCC1 in colorectal carcinogenesis

Adam P, Salée C, Stepniak M, Loly J-P, Vieujean S, Reenaers C, Fonzé F, Massot C, Bletard N, Coimbra Marques C, Decker E, Delvenne P, Louis E & Meuwis M-A

NKCC1 is a Na-K-Cl cotransporter localized at the cell membrane and in the cytoplasm, predominantly expressed in cells involved in fluid secretion. NKCC1 is overexpressed in pre-tumoral and tumoral lesions of colitis-associated cancer and colorectal cancer (CRC). Moreover, NKCC1 expression has been associated with survival outcomes of CRC patients. However, its function in colorectal carcinogenesis remains underexplored. NKCC1 is associated with intestinal stem cells and has been recently identified in a colorectal cancer stem cell signature. These colorectal cancer stem cells have been specifically associated with poor cancer prognosis, treatment resistance, and deregulated pathways, including altered ROS levels and oxidative stress responses. We tested the inhibition of NKCC1 ion transport activity using bumetanide under oxidative stress (1.5 mM H2O2) in CRC cell lines which resulted in and enhanced p70 S6 kinase phosphorylation and a 4.76x increase cell mortality (viability of 35.2% for control versus 7.5% for bumetanide, p = 0.0224). These results suggest that NKCC1 might support tumor cell survival under oxidative stress, potentially involving the mTORC1 pathway. By silencing NKCC1 via shRNA in CRC cell lines, we observed a decrease in cell proliferation and an increased expression of cancer stem cell markers (LGR5, CD44, ALDH1A1). We are currently investigating transcriptomic changes upon NKCC1 silencing in HT29 cells (bulk RNA sequencing). We will use the same strategy on patient-derived intestinal tumoroids and organoids for implementing NKCC1 characterization in tumor cells and normal phenotype epithelial intestinal cells, respectively. Both strategies of inhibition should clarify why NKCC1 is upregulated throughout the tumor transformation sequence, starting from early lesions (dysplasia) to advanced colorectal tumors and disclose its potential impact in colorectal carcinogenesis.

Poster 2

Ning An

Ning.An@uliege.be

Laboratory of Cancer Signaling
Cancer

tRNA wobble editing dictates ferroptosis sensitivity in lung cancer

Reprogramming of mRNA translation is central to cancer development, mediating cancer cell adaptation and supporting tumor progression. tRNAs are highly modified molecules that are essential to correctly translate mRNAs into proteins. Recently, the importance of tRNA-modifying enzymes in promoting cancer development and therapy resistance through codon-specific translation reprogramming has been uncovered. tRNA-specific adenosine deaminase 2 (ADAT2) is an evolutionarily conserved enzyme that catalyses the conversion of adenosine to inosine at the wobble position of tRNAs (A34). Here, we found that ADAT2 is up-regulated in human lung cancers and is essential for the growth of lung cancer cells and xenograft tumors. Importantly, genetic deletion of Adat2 in mice lungs strongly impairs tumor development in a KrasG12D/+ model of spontaneous lung cancer. Using a combination of proteomics, polysome profiling and ribosome sequencing, we demonstrate that ADAT2 depletion directly alters the translation of a specific subset of metabolic enzymes that are enriched in C-ending and G-ending (NNC and NNG) codons. As a result, ADAT2 loss in lung cancer cells leads to impaired glucose and glutamine metabolism, accumulation of reactive oxygen species and rewiring of lipid metabolism. In addition to global metabolic changes, we demonstrate that ADAT2 directly regulates the translation of the ferroptosis suppressor protein 1 (FSP1). Accordingly, ADAT2 knockdown decreases FSP1 expression, triggers ferroptosis activation and sensitizes lung cancer cells to GPX4 inhibition. Finally, we show that an ADAT2 translational signature is enriched in KEAP1-mutated lung tumors and correlates with poor outcome in lung cancer patients. Taken together, our data uncover the importance of tRNA wobble editing in controlling cellular homeostasis in lung cancer and highlight new metabolic vulnerabilities to be exploited for future therapies.

Poster 3

Esther Arpigny

esther.arpigny@uliege.be

Laboratory of Connective Tissue Biology (LCTB)
Cancer

Identification of the Roles of ADAMTS12 Secreted by Stellate Cells during Tumor Progression in Liver Cancer

Arpigny E, Legagneux V, Théret N, Colige A

We identified ADAMTS12 as a metalloproteinase strongly associated with recurrence risk in Hepatocellular Carcinoma. We also determined that ADAMTS12 is specifically expressed by activated HSC (hepatic stellate cells) present in the tumor and that siRNA-mediated repression of ADAMTS12 affects the transcriptome of LX-2 cells (a human HSC line). In this study, we aim to identify how ADAMTS12 regulates the phenotype of HSCs and modulates their dialogue with cancer cells. We first generated a relevant cell model. Using Crispr/Cas9 technology, we generated LX-2 cells in which the endogenous ADAMTS12 activity is suppressed (LX-2 KO). These cells were then engineered to conditionally express recombinant ADAMTS12 in the presence of doxycycline. LX-2 cells, expressing or not ADAMTS12, were characterized by RNA-Seq. Relevant pathways involved in tumor microenvironment remodeling, endoplasmic reticulum stress response and lipid metabolism were identified as genes regulated by ADAMTS12 expression. As compared to controls, LX-2 cells expressing ADAMTS12 are characterized by an increased number of cytoplasmic lipid droplets, in line with the RNA-Seq data. Furthermore, hepatocellular carcinoma (HepG2) cells exposed to medium conditioned by LX-2 cells overexpressing ADAMTS12 also show an increase in the amount of lipid droplets, suggesting an involvement of ADAMTS12 in the regulation of lipid metabolism. For in vivo studies, we first generated MMTV-PyMT mice (which spontaneously develop mammary tumors) which are further deficient in ADAMTS12 (PyMT-TS12-KO). As compared to MMTV-PyMT wild type, mice PyMT-ADAMTS12-KO develop smaller mammary tumors. Based on these data showing that ADAMTS12 is involved in the regulation of lipid homeostasis and can modulate tumour growth, ongoing researches are trying to verify whether these two effects are functionally linked. We also aim to decipher the involved mechanisms by trying to identify factors whose cleavage by ADAMTS12 could be involved in the observed regulations.

Poster 4

Louis Baudin

louis.baudin@uliege.be

Laboratory of Biology of Tumor and Development
Cancer

Vascular, immune and extracellular matrix remodeling in lymph node (pre-)metastatic niche: new insights into nodal metastases

Baudin L, Zanella L, Josse T, Blacher S, Kridelka F, Noel A

Several cancers, including cervical cancers and melanomas, are known to metastasize primarily to lymph nodes (LN). Recent data demonstrate that LN metastases can seed to distant organs, revealing that LN are worth considering as potential therapeutic target to prevent distant disease and death. Through a complete analysis of 68 patient-derived samples, we confirmed here the dialogue between the primary cervical neoplasm and the pelvic sentinel LN in early cervical cancer at multiple levels. The access to a large cohort composed of 189 patients suffering of advanced cervical cancer allows an in-depth investigation, combining IHC and computational analysis, of the implication of vascular structures, immunity and extracellular matrix proteins in the elaboration of a permissive niche for metastatic colonization. Thanks to the associated clinical data including the patient survival, the relapse and the relapse site, we are trying to generate a robust and original protein signature both at the remodeled-LN and the primary tumor level. A special focus on metastatic lymph node led us to the identification of a local remodeling around the nodal metastasis. Indeed, a dense aSMA-positive fibroblastic coat, as well as an increased FOXP3-positive cell number close to the metastatic border were identified, suggesting the elaboration of a local and immunotolerant peri-metastatic environment. Deciphering such mechanisms is crucial in order to understand the paradoxal way in which tumor cells are able to survive and settle in a site responsible of the immune response, namely the lymph node. Finally, a deeper understanding of the tumor/lymph node dialogue and the identification of nodal markers predicting the risk for distant extension are key prognosis variables and so, are worth considering for patients suffering malignancies that disseminate through the lymphatic vasculature.

Poster 5

Chloé Beaudou

cbeaudou@uliege.be

Laboratory of Neuroendocrinology
Neuroscience

Role of kisspeptin and GnRH in memory formation and retention

Beaudou C, Bakker J

Kisspeptin is a neuropeptide known for its crucial role in the hypothalamic-pituitary-gonadal axis (HPG) that controls reproductive function. The populations of kisspeptin neurons are located in the hypothalamus, more precisely in the anteroventral periventricular area (AVPV) and arcuate nucleus (ARC). Both modulates the pulsatile release of GnRH, responsible for the pre-ovulatory peak of LH. Interestingly, the kisspeptin receptor, GPR54, is present in extra-hypothalamic areas such as the cortex and hippocampus among others. These observations suggest that kisspeptin and GPR54 may play a role in non-reproductive functions such as cognition, mood or social behaviors. Therefore, we propose to study the role of kisspeptine and GnRH in mouse memory. First, the injection of a retrograde viral vector in Kiss::Cre mice show that the AVPV kisspeptin neurons population project to the dorsal region CA1 of the hippocampus. Then, we administered the female wild-type mice with kisspeptine or GnRH and performed on these mice the new object recognition test (NOR) which allows to evaluate memory retention. Administration of these two drugs showed a significant increase in memory performance in these mice. Finally, by using chemogenetic method, we injected a viral vector allowing the expression of hM4Di in the AVPV nucleus in Kiss mice:Cre to specifically inhibit the AVPV kisspeptin neurons. These mice showed significantly decreased memory retention performance compared to the controls. Together, these results suggest that kisspeptine and the GnRH play a role in memory formation and retention.

Poster 6

Kübra Bekar

kubra.bekar@uliege.be

Laboratory of Molecular Angiogenesis
Cancer

Insights Into The Role Of Endothelial Extracellular Vesicles In Pre-Metastatic Niche Formation And Metastasis In Breast Cancer

Bekar Â, Struman I
Laboratory of Molecular Angiogenesis, GIGA Research Centre, University of Liège, Liège Belgium.

Breast cancer (BC) is one of the most common cancers worldwide. While it can be treated, metastatic BC remains incurable. Previously, our research group demonstrated that endothelial cell-derived extracellular vesicles (EVs) enriched in miR-142-5p, miR-183-5p, and miR-222-3p participate in the polarization of macrophages towards an M2-like phenotype, thus promoting tumour growth in a BC mouse model (Njock et al., 2022, JEV). However, the impact of these microRNAs on metastasis remains unknown. A crucial step during metastasis is the formation of a pre-metastatic niche (PMN), which is notably initiated by EVs. With this project, we aim to unravel the impact of these microRNAs on PMN formation and metastasis in vitro and in vivo. In this study, we isolated EVs from mouse endothelial cell lines, characterized them using Western blotting (WB) and Nanosight Tracking Analysis (NTA). Next, we evaluated their incorporation into macrophages and fibroblasts, which are two major cell types in PMN formation, using confocal microscopy. Afterwards, we electroporated the EVs with the three microRNAs and added them to macrophages and fibroblasts. To determine the effects of these EVs on cell differentiation, we analysed the expression by qPCR of several genes known to be involved in PMN formation. Our data showed that endothelial EVs were successfully incorporated into the recipient cells. Our qPCR results demonstrated that microRNA-enriched EVs upregulate the expression of pro-tumorigenic genes Csf3, Cxcl1, Col3a1, Il-1_, and Ccl3. Currently, we are further investigating the role of microRNA-enriched endothelial EVs in PMN formation in a 4T1-BC mouse model. Our current findings reveal a previously unrecognized role of microRNA-enriched endothelial EVs on macrophage and fibroblast differentiation in vitro. These preliminary findings reveal a potential role of endothelial EVs in PMN formation, offering avenues for novel approaches in BC treatment.

Poster 7

Laëtitia Bernet

laetitia.bernet@uliege.be

Laboratory of Biology of Tumor and Development
Cancer

Exploring the Impact of Chemotherapy Dosing on Ovarian Function and Long-Term Fertility in Peripubertal Mice

Bernet L, Bindels J, Lowette C, Munaut C

Female children, adolescents, and young adults (CAYA) surviving cancer treated with high doses of chemotherapy have an elevated risk of ovarian failure and infertility. While fertility preservation is highly recommended for post-pubertal girls receiving high doses, its necessity for prepubertal girls and those exposed to lower doses remains debatable, highlighting the need for more research on timing and dosage for fertility preservation. In this study, peripubertal female C57BL/6 mice aged 4 weeks received either a single injection of cyclophosphamide (Cyp) and busulfan (Bus) at doses of 12/1.2 mg/kg or 120/12 mg/kg, or 6 injections over two weeks at doses of 7.5/0.75 mg/kg, 15/1.5 mg/kg, 37.5/3.75 mg/kg, 75/7.5 mg/kg. We assessed follicular density and function in ovaries harvested 24 h or seven days after the last injection. Fertility was evaluated by monitoring the estrous cycle for 3 weeks and through mating experiments. The results show that both chemotherapy protocols significantly reduced ovarian reserve in peripubertal mice. Moreover, mice receiving multiple injections displayed a significantly higher percentage of _H2AX-positive follicles, indicating DNA damage, compared to controls, whereas mice receiving a single high-dose injection showed no significant differences from controls. At reproductive age, a single injection of 120/12 mg/kg Cyp/Bus did not impact fertility, while multiple injections of 75/7.5 mg/kg Cyp/Bus led to complete impairment of estrous cycles and fertility. This mouse model developed with six chemotherapy injections over two weeks, serves as a valuable tool for studying long-term chemotherapy effects and mimics the multiple administrations seen in clinical settings. Future experiments will explore ovarian cryopreservation and transplantation post-chemotherapy in young mice, using this model.

Poster 8

Jules Bindels

jules.bindels@uliege.be

Laboratory of tumor and development biology
Cancer

Inhibition of the AKT/mTOR Pathway using Rapamycin Enhances Follicle Survival and Fertility Preservation in Ovarian Tissue Transplantation

Bindels J, Squatrito M, Bernet L, Nisolle M, Munaut C
Laboratory of Biology of Tumor and Development, GIGA-Cancer, Université de Liège
Department of Obstetrics and Gynecology, Hôpital de la Citadelle, Université de Liège

At present, the sole fertility preservation method available for prepubertal patients or those needing immediate treatment for malignancies is ovarian tissue cryopreservation followed by autotransplantation (OTCTP). This technique, which has led to over 200 live births, involves autotransplantation of slow-frozen (SF) and thawed tissue once patients are in remission and wish to conceive. However, significant follicular loss due to premature follicle activation post-transplantation remains a challenge.
To improve follicular survival during OTCTP, we investigated the inhibition of the Akt/mTOR pathway using rapamycin, a known mTOR inhibitor. We used a murine heterotopic ovarian transplantation model to evaluate whether rapamycin could improve fertility preservation. First, we assessed the impact of rapamycin on OTCTP-induced follicle activation after transplantation under the kidney capsule of mice ovaries for 3 weeks (fresh, SF, or SF with rapamycin). We performed immunohistochemistry (IHC) assisted follicle quantification and analysed follicle proliferation and activation using IHC. Next, we tested rapamycin's efficacy in an orthotopic transplantation model, where mice were subjected to unilateral oophorectomy, SF of ovaries with/without rapamycin, and chemical disabling of the remaining ovary. Post-transplantation, these mice were mated for 4 months to evaluate pregnancy outcomes.
IHC analyses of ovarian grafts revealed that rapamycin effectively counteracted OTCTP-induced follicle proliferation and Akt/mTOR pathway activation. In the orthotopic model, mice whose ovaries were frozen with rapamycin exhibited gave birth to more pups with a greater live birth rate compared to controls.
Our results indicate that adding the mTOR inhibitor rapamycin during OTCTP can transiently maintain primordial follicles in a quiescent state, significantly enhancing fertility restoration in mice. This approach could potentially improve clinical outcomes for patients undergoing OTCTP.

Poster 9

Paradeisios Alexandros Boulakis

paradeisios.boulakis@uliege.be

Laboratory of Physiology of Cognition
Neuroscience

Mind-blanking frequency alters across different autonomic arousal states

Boulakis PA, Simos NJ, Zoi S, Mortaheb S, Raimondo F, Demertzi A
Physiology of Cognition Lab, GIGA CRC Human Imaging, University of Liege
Sleep and Chronobiology, GIGA CRC Human Imaging, University of Liege
Institute of Neuroscience and Medicine (INM-7: Brain and Behaviour), Research Center Julich

In this registered report (https://osf.io/nfcvu/), we examined whether the inability to report any immediate content, termed mind-blanking (MB), can result from the manipulation of a person's current physiological state. In a repeated measures design, 26 participants performed an experience-sampling task (40 trials; inter-probe interval = 110s) under different autonomic arousal conditions while multimodal brain-body recordings were taking place. During baseline measurements, participants reported their thoughts by opting across a) sensations, b) mind-wandering, and c) blank. The protocol was repeated in counter-balanced order in a high-arousal (post high-intensity exercise) and a low-arousal condition (post 8-hour sleep deprivation). According to our hypothesis, MB reports were increased in the low arousal condition compared to baseline (b=-.794, pFDR<.000) and high arousal (b=-.968, pFDR<.000). We found partial evidence for a temporary increase of blanks during the high arousal condition that did not last across the experience-sampling session. Utilizing a machine learning approach, we further showed that a balanced random-forest classifier can decode MB reports from a brain-body matrix above chance level (balanced accuracy = .66), and outperforms classifiers trained solely on brain (balanced accuracy = .64) or body (balanced accuracy = .61) features. This result was maintained when the classifier was trained on separate arousal conditions, and when considering different analysis windows. Our results suggest that MB is an arousal-modulated mental state and that peripheral physiology carries unique information about MB that cannot be encapsulated solely in brain activity. Overall, we show that an embodied approach provides greater explanatory power in understanding MB.

Poster 10

Sharon Brembati

sharon.brembati@uliege.be

Cancer Stemness Laboratory
Cancer

Characterization of the trnas of lung cancer stem cells

Brembati S, Rapino F

Lung cancer remains a major cause of cancer-related deaths worldwide with unfavorable prognosis mainly due to the late stage of disease at presentation. Malignant phenotype in lung cancer is sustained by transformed alveolar epithelial type II (ATII) lung cancer stem cells (CSCs) that have the potential of initiating lung cancer formation, mediate metastasis formation and support resistance to therapy. A growing amount of evidence suggest that mRNA translation regulation and tRNA modifications play a key role in supporting cancer establishment. Therefore, we postulate that the expression of a specific signature of tRNA enzymes permits the establishment lung cancer stem cells by sustaining proteome rewiring upon KRAS oncogenic transformation. To investigate this hypothesis, we optimize human healthy lung spheroids and KRASG12S lung cancer spheroids cultures to increase the expression of stemness markers (CD133/CD44), anchor-free survival and EMT markers. We then performed a set of drop-out CRISPR-Cas9 pool screens of all the known human tRNA enzymes to identify new mRNA translation modulators of lung CSC. Through this approach, we highlighted five tRNA enzymes responsible for three different types of tRNA modifications that specifically impact stem-like proprieties of KRASG12S lung cancer cells but not of healthy cells. To validate our targets, we depleted the tRNA enzymes by shRNA and quantified CSC fitness by following spheroid formation and the capability of cells to undergo EMT. Our preliminary results indicate that the loss of our targets impacts lung CSC fitness. Currently, we are generating ribo-seq and proteomic datasets of KRASG12S lung cancer spheroids depleted or not of our targets to better characterize the impact of our targets on lung CSC proteome establishment.

Poster 11

Maureen Cambier

mcambier@uliege.be

Laboratory of Molecular Angiogenesis
Cancer

Unraveling the significance of exosomal immune checkpoint protein PVR/CD155 in Lung Carcinoma progression

Cambier M, Remacle C, Beyens J, Herkenne S, Henket M, Gillet H, Polese B, Schyns J,, Sibille A, Louis R, Guiot J, Struman I.

Immunotherapy has revolutionized cancer treatment, but understanding the resistance to this treatment is crucial for improving outcomes. In this study, we investigate the role of extracellular vesicles (EVs) in immune escape through immune checkpoint protein (ICP) expression, focusing on the axis PVR/CD155. We developed a new methodology that identifies several ICPs on the surface of EVs in single-liquid biopsies. We analyzed the expression of several ICPs identified through literature screening and TCGA data analysis. Plasma samples from healthy individuals and cancer patients undergoing immunotherapy were collected. EVs were isolated and characterized using ultracentrifugation. ICPs on EV surfaces were identified via MAGPIX analysis. Knockdowns were generated with siRNA and CRISPR-Cas9 and studied with functional assays. Analysis of TCGA data revealed high levels of PVR/CD155 in lung cancer. EV-associated PVR/CD155 levels were elevated in patient samples. Furthermore, its expression was even more abundant during immunotherapy in non-responders, suggesting an emergence of resistance. We studied ICP expression in several human and mouse models and discovered an enrichment of PVR in EVs. Knockdown of PVR/CD155 cells presented reduced migration and proliferation rates in human and mouse models. Our study introduces a novel method for characterizing circulating ICPs-EVs in lung cancer patients, with implications for patient monitoring and therapeutic target discovery. Further investigation of PVR/CD155+EV in coculture systems with immune cells will deepen understanding of its role in immunotherapy resistance.

Poster 12

Manon Charlet

m.charlet@uliege.be

Molecular Regulation of Neurogenesis
Neuroscience

Alcohol binge-drinking during adolescence impairs behaviors in adulthood by modulating mTORC1 activity in the prefrontal cortex

Charlet-Briart M, Oskera L, Nguyen L, Laguesse S

During adolescence, the brain undergoes intense maturation in the frontal areas. The immaturity of the prefrontal cortex (PFC) in adolescents is associated with heightened vulnerability to the deleterious effects of drugs. Indeed, excessive alcohol consumption may interfere with the ongoing maturation of frontal brain circuits, leading to long-lasting consequences in PFC structure and function. Clinical studies have shown that excessive adolescent alcohol exposure (AAE) significantly increases the risk of developing psychiatric disorders later in life, including addiction. However, the mechanism by which alcohol perturbs the maturation of the adolescent PFC is still poorly understood. The “mechanistic target of rapamycin complex 1" (mTORC1) is a master regulator of translation, which promotes alcohol-dependent neuroadaptations in the adult nucleus accumbens and the development of alcohol use disorders (AUD). Here, we demonstrate that voluntary binge-drinking of alcohol during adolescence activates mTORC1 in the PFC, especially in the layer V pyramidal tract neurons of the prelimbic and infralimbic cortex, whereas this is not the case in the motor cortex. Similar results were observed in males and females. Moreover, counteracting the alcohol-induced increased mTORC1 activity with rapamycin injections during adolescence rescued the anxiety-like phenotype as well as the excessive alcohol consumption in adulthood. Currently, we are (i) deciphering the neuronal-type specificity of the alcohol-induced modulation of mTORC1, (ii) unveiling the structural and functional consequences of increased mTORC1, and (iii) identifying the mRNAs whose translation is induced by AAE in a mTORC1-dependent manner.

Poster 13

Maëlle Charonitis

maelle.charonitis@uliege.be

CRC Human Imaging
Neuroscience

Exploring Cognitive Load's Effects on Post-Stroke Mental Fatigue

Charonitis M, Maquet P, Collette F.
GIGA-CRC Human Imaging, University of Liège, Liège, Belgium
Department of Neurology, Liège University Hospital, Liège, Belgium

Mental fatigue is common among stroke survivors, manifesting as exhaustion after prolonged mental effort. However, its impact on cognition is not well understood. This study examines how induced mental fatigue affects cognitive performance in stroke patients compared to healthy controls (HC) and whether stroke patients show a greater need for rest than HC. Twenty-three ischemic stroke patients and 19 matched HC underwent a mental fatigue induction task, manipulating cognitive load by adjusting task duration and stimulus presentation times. The high cognitive load (HCL) condition had an extended task duration (32min) and reduced stimulus presentation time compared to the low cognitive load (LCL) condition (16min). Delta scores, reflecting the difference in accuracy between last and first blocks, were computed. Participants rated their fatigue levels pre- and post-task using a visual analog scale and their rest propensity in daily life through the BFS questionnaire. Linear mixed models were used to investigate the effects of group, time (only for subjective fatigue), and cognitive load on delta scores and fatigue levels. Group differences in rest propensity were examined using a Mann-Whitney test. Analyses revealed that subjective fatigue levels increased over time-on-task ($p=.014$) and were higher in HCL compared to LCL ($p<.001$). However, no group difference was observed. Performance decline was more pronounced among stroke patients in HCL compared to both stroke ($p=.027$) and HC ($p<.001$) in LCL. Stroke patients reported a greater need for rest following intellectual activities compared to HC at BFS ($p=.013$). Stroke patients showed greater susceptibility to mental fatigue, as evidenced by their decline in accuracy under higher cognitive load and increased need for rest following mental activities. However, this finding contradicts their reported fatigue levels during the task. Compensatory brain mechanisms and motivational factors may contribute to this discrepancy.

Poster 14

Sanket Chiplunkar

SSChiplunkar@doct.uliege.be

Laboratory of Human Genetics/Medical Oncology
Cancer

Systemic Eosinophil Depletion (S.E.D.) reduced the efficacy of response to anti-PD1 checkpoint blocking immunotherapy in a TNBC murine model

Chiplunkar S, Boulet D, Gilon M, Thiry J, Josse C, Jerusalem G, Bours V

Breast cancer (BC) is among the most common types of cancer and remains a significant cause of cancer-related mortality worldwide. Among several BC subtypes, triple negative (TNBC) is the most difficult to treat with worst prognosis. Currently, aggressive TNBC cases are routinely treated by checkpoint blocking immunotherapies. Unfortunately, not all patients respond equally well to this treatment. Eosinophils are one of the immune cell-types known to be involved in this treatment response. But unlike many other immune cells, eosinophils are perhaps the least studied with paucity of knowledge regarding their exact roles due to a very limited number of pre-clinical investigations. Using murine models, my project is investigating eosinophils in the BC progression and therapeutic response. The employed experimental strategy includes tumor cell-injection in the mammary fat-pad of mice and comparative analysis of tumorigenic progression between Systemic Eosinophil Depletion (SED) group and a control group with unaltered eosinophil count. So far, our results suggest no role for eosinophils in the development of primary mammary-tumor and secondary lung metastasis. However, in the same pre-clinical TNBC model (4T1 in Balb/C), we did notice statistically significant primary tumor shrinkage in the control group compared to the SED group upon therapeutic anti-PD1 checkpoint blockade, highlighting the eosinophil involvement in response to this immunotherapy. In future, we plan to confirm these results in a second BC model with tail-vein injected EO771 cells in C57BL/6 to investigate SED impact on lung metastasis (ultimate cause of death from BC) and therapeutic response to chemotherapy and checkpoint blocking immunotherapy.

Poster 15

Isabelle Dias Da Silva

i.diasdasilva@uliege.be

Laboratory of Tumors, Biology and Development
Cancer

Unraveling the Transcriptional Landscape of Estradiol, Progesterone, and Bazedoxifene in Human Endometrial PDX Models

Dias Da Silva I, Wuidar V, Staumont B, Nisolle M, Métivier R, Péqueux C
Laboratory of Biology, Tumor and Development, Giga-Cancer, University of Liège
Biomechanics Research Unit, GIGA In Silico Medicine, University of Liège
Gynecology department, Giga-Cancer, CHU and University of Liège
Institute of Genetics and Development of Rennes (IGDR), UMR 6290, CNRS, University of Rennes, Rennes, France

Estrogens are widely used for menopause treatment, and selective estrogen receptor modulators (SERM) are developed for endocrine therapy of breast cancer. The endometrium, being highly estrogen-sensitive, is a main target of these treatments. Menopause hormone treatment (MHT) with estrogen-only preparations increase endometrial cancer risk, prevented by adding a progestogen. However, combining estrogen and progestogen increases breast cancer risk. Bazedoxifene (BZA), a third-generation SERM, offers a promising alternative by antagonizing estrogenic effects in both breast and endometrial tissues. This study investigates the transcriptomic and functional effects of Estradiol (E2) alone or in combination with Progesterone (P4) or BZA in patient-derived xenograft (PDX) models of human endometrium. Preliminary RNA sequencing data show that BZA significantly modifies gene expression, countering E2-induced changes, while P4 influences cell proliferation pathways. Using 3D organoid culture, RNA sequencing and immunofluorescent staining, we elucidate the molecular mechanisms of these treatments on human endometrium.
Our results reveal distinct transcriptional profiles and cellular effects of these treatments, highlighting BZA's potential as a safer alternative. These insights pave the way for developing next-generation hormonal therapies, optimizing efficacy while minimizing risks, and highlighting the importance of detailed molecular profiling in guiding safer treatment strategies.

Poster 16

Bianca E Silva

b.silva@uliege.be

Laboratory of Hematology
Immunobiology

IFN-_ predisposes acute myeloid leukemia to therapy resistance

E Silva B, Daubry A, Faville C, Jassin M, Baron F, Ehx G

Acute myeloid leukemia (AML) patients frequently relapse following frontline therapies such as high-intensity chemotherapy and hypomethylating agents. Currently, the mechanisms underlying this therapy resistance remain elusive. We recently discovered that AML blasts present MHC peptides recognized by T cells at diagnosis, leading to their activation and subsequent secretion of cytokines. While such a response might help eradicate leukemia, we hypothesized that pro-inflammatory cytokines secreted by immune cells might also contribute to therapy resistance. Therefore, we performed large-scale transcriptomic analyses comparing blasts obtained at diagnosis from patients who either responded or did not respond to conventional cytarabine + anthracycline therapy. Our analyses revealed an upregulation of IFN-_ signaling signatures in patients resistant to therapy. Similar signatures were observed in patients resistant to the hypomethylating agent 5-azacytidine. Consequently, patients expressing high IFN-_ signaling scores at diagnosis had lower survival rates. Additionally, treating multiple AML cell lines with IFN-_ in vitro significantly decreased the cytotoxic activity of high doses of chemotherapeutic agents; treated cells exhibited faster proliferation rates following chemotherapy exposure than untreated cells, suggesting that IFN-_ signaling may accelerate relapse in patients. In conclusion, our findings suggest that inhibiting IFN-_ signaling might help overcome therapy resistance in AML.

Poster 17

Florence Evrard

fevrard@uliege.be

Laboratory of Neuroendocrinology
Neuroscience

Social behaviour in mice of both sexes after ablation of PVN nNOS neurons

Evrard F, Bakker J
Laboratory of Neuroendocrinology, GIGA-Neurosciences, University of Liege

Social behaviour, including sexual and aggressive behaviour, is both innate and adaptive, relying on complex neural circuits with significant sex differences. Nitric oxide, synthesized by neuronal nitric oxide synthase (nNOS), may play a crucial role in these circuits, as suggested by our previous experiments. Here, we investigated the effects of specific ablation of nNOS neurons in the hypothalamic paraventricular nucleus (PVN) on the social behaviour of male and female mice using an AAV-mediated Cre-dependent caspase system and a battery of tests. Our results show that PVN-nNOS-Del mice of both sexes show a loss of preference for the opposite sex. In males, this is accompanied by improved sexual performance: males engaged in sexual activity more frequently, initiated mounting and intromissions faster and performed more intromissions compared to controls treated with AAV lacking a caspase-encoding transgene. In contrast, female sexual behaviour was largely unaffected. Assessment of aggression was difficult due to low levels of aggression despite a protocol to increase it. In addition, PVN-nNOS-Del mice showed no changes in sociability or preference for social novelty. Despite the key role of the PVN in anxiety regulation, PVN-nNOS-Del mice showed little to no reduction in anxiety levels. These results highlight the selective role of PVN nNOS neurons in the regulation of sexual preference and sexual behaviour in males, with minimal effects on female sexual behaviour, sociability and anxiety. This highlights the nuanced and sex-specific functions of nNOS neurons within the PVN. The findings support the hypothesis that PVN nNOS neurons may influence sexual preference by influencing neural circuits involved in attraction to the opposite sex, and may play a role in controlling inhibitory mechanisms associated with male sexual activity. Further research is needed to investigate their interactions with other neural circuits involved in social behaviour.

Poster 18

Charline Faville

Charline.Faville@uliege.be

Laboratory of Hematology
Cancer

Optimization of an MHC-I immunoprecipitation protocol for mass spectrometry analyses of persister-specific antigens in acute myeloid leukemia

Faville C, E Silva B, Vialnova Mana L, Cobraiville G, De Voeght A, Baron F, Fillet M, Baiwir D, Mazzucchelli G, Willems L, Ehx G.

Hematology Research Unit, Groupe Interdisciplinaire de Génoprotéomique Appliquée (GIGA), University of Liège
Cellular and Molecular Epigenetics Research Unit, Groupe Interdisciplinaire de Génoprotéomique Appliquée (GIGA), University of Liège
Laboratory for the Analysis of Medicines, Department of Pharmaceutical Sciences, CIRM, University of Liège
Department of Medicine, Division of Hematology, CHU of Liège
Proteomics Facility, Groupe Interdisciplinaire de Génoprotéomique Appliquée (GIGA), University of Liège

Acute myeloid leukemia (AML) is the most frequent and lethal leukemia among adults. Chemotherapy (cytarabine (Ara-C)) is the first-line treatment option and leads to high remission rates (75%). However, most patients eventually relapse. Relapse is mediated by persisters (i.e., cells surviving the treatment) that present a vast transcriptomic reprogramming characterized by the expression of stress responses, such as senescence and diapause. Since we previously showed that alterations in the transcriptome are reflected in the immunopeptidome (set of MHC-I-associated peptides (MAP)), we hypothesize that persisters present specific and immunogenic MAP deriving from stress responses. Therefore, we aim to identify such MAP to design a peptide vaccine targeting these cells and preventing relapse. Mass spectrometry (MS) on immunoprecipitated MHC-I molecules and MAP is the only method to explore the immunopeptidome. Preliminary analyses showed that few MHC-I molecules could be isolated from AML cell lines. Accordingly, few MAP (31) were identified by MS. Therefore, we sought to improve the yield of our isolation method and found that performing eight elutions instead of two dramatically increased the amount of collected MHC-I. We also found that using next-generation trapped ion mobility time-of-flight (timsTOF) MS dramatically (~300-fold) increased the number of identified MAP. After bioinformatic filtering of peptides based on their size distribution and predicted binding to MHC-I molecules, a clear enrichment of MHC-I epitopes was observed. Identified MAP were also radically different (~20% overlap) and had greater GRAVY indexes than others identified previously in the same cell line with Orbitrap MS, suggesting that timsTOF enables the discovery of previously unreported MAP, due to its capacity to capture more hydrophobic MAP. In conclusion, our MS protocol is operational and will allow us to identify enough MAP to find good candidates for designing the vaccine.

Poster 19

Jeremy Gardette

j.gardette@uliege.be

CRC Human Imaging
Neuroscience

Individual differences in anterograde memory for details relate to the posterior hippocampus volume

Gardette J, Besson G, Baillet M, Rizzolo L, Narbutas J, Van Egroo M, Chylinski D, Maquet P, Salmon E, Vandewalle G, Collette F, Bastin C

In recent years, there has been a growing interest in individual differences in autobiographical memory. The ability to recall details from personal past events was found to correlate with the volume of specific subfields of the hippocampus in healthy adults. Although the posterior region of the hippocampus is believed to process detailed memory representations independently of the memory's age, little is known about individual differences in the ability to recall newly encoded events in detail, and how these differences relate to hippocampal subregions. In this preregistered study, we scored the story recalls from 89 healthy middle-aged participants with a newly designed method that allows to distinguish information recalled in detail from gist recall (i.e., when only the general idea is recalled). Between the immediate and the delayed recall (~ 20 minutes), detailed information was transformed into gists, which is in line with recent evidence that gists can emerge rapidly after a new experience. In addition, we segmented the anterior and posterior hippocampal subfields CA1, CA2/3, dentate gyrus, and subiculum from high-resolution structural MRI. As predicted, the volume of the posterior hippocampus was positively correlated with the detail score but not with the gist score, yet this effect was restricted to the right hemisphere. We also observed trends towards associations between the detail score and specific subfields of the right posterior hippocampus, but none survived statistical correction. Finally, we found no evidence for the expected age-related increase in the use of gists over details. Taken together, these results suggest that the posterior hippocampus supports detail memory in the recall of both remote and newly acquired memories.

Poster 20

Alexia Genco

alexia.genco@uliege.be

Laboratory of Molecular Analysis of Gene Expression
Cancer

Deciphering the molecular properties of the putative RNA-binding protein TFIP11 in regulating alternative splicing

Obeid A, Galvan B, Flasse L, Pendeville H, Mottet D

RNA-binding proteins (RBPs) are a class of proteins that regulate the metabolism of RNAs throughout their life cycle. Many RBPs can be cell-, tissue- or condition specific and are capable of regulating various molecular processes such as alternative splicing of messenger RNAs which is essential for the proper expression of multiple proteins. These proteins possess domains essential for interaction with the target RNA called RNA binding domains. Typically a RBP has multiple RBDs. Here, we are studying the TFIP11 protein, a spliceosomal protein that possesses two putative RBDs; a G-patch domain at its N-terminal region and a dsRBD at its C-terminal region. The laboratory has already demonstrated the importance of TFIP11 in regulating spliceosome assembly and activation but little is known about the functionality of its RNA-binding modules, nor about the molecular mechanisms governing its role in regulating alternative splicing. Our current study focuses on the importance of TFIP11 domains for the formation of splicing complexes, as well as their importance for protein-RNA interaction, as these protein/protein and protein/RNA interactions can govern splicing decisions that ultimately impact the alternative splicing of a set of essential genes.

Poster 21

Zoé Gendebien

zoe.gendebien@chuliege.be

Laboratory of Rheumatology
Immunobiology

Coexpression and infiltration of endoplasmic reticulum stress chaperone proteins together with CD34+ fibroblast-like synoviocytes in human inflamed synovial membranes

Gendebien Z, Hego A, Poulet C, Deroyer C, Paulissen G, Hemon P, Marec N, Durez P, Bianchi E, Delvenne P, Ribbens C & de Seny D

Eleven endoplasmic reticulum (ER) stress chaperone proteins (BiP, HYOU1, MANF, PDIA4, GANAB, HSP90B1, TXNDC5, DNAJB11, LMAN1, ERP29, and CALR) were identified by mass spectrometry in synovial membranes from patients with osteoarthritis (OA), chronic pyrophosphate arthropathy (CPPA), and rheumatoid arthritis (RA). These proteins were highly correlated with an inflammatory histological score based on hyperplasia and leukocyte infiltration. In addition to their role in protein folding, these ER chaperones can enhance inflammation and immunogenicity under ER stress. Our previous work has shown that these proteins are restricted to the lining layer during mild inflammation and infiltrate the synovium during severe inflammation. The differential expression of some ER stress proteins was confirmed using the CIOA mouse model. In vitro, these were expressed by fibroblast-like synoviocytes (FLS) under basal conditions, but were upregulated and even secreted after ER stress, after a profibrotic or after a proinflammatory induction. This study aims to precisely localize these eleven ER stress proteins within the cell subpopulations of inflamed synovial membranes. Imaging mass cytometry was performed on formalin-fixed paraffin-embedded synovial membranes obtained from patients with OA (n=5), naive RA (n=1), late RA (n=3), and undifferentiated arthritis (n=4). Tissue sections were conjugated with metal antibodies recognizing synovial cells (FLS, immune cells, ...), followed by laser desorption and time-of-flight mass spectrometry to detect and quantify the spatial distribution of multiple proteins simultaneously. Our data show that ER stress proteins co-express with CD55, characterizing FLS from the lining when the inflammation is mild, but infiltrate the sublining together with CD34+ cells (sublining FLS) when the inflammation increases. Interestingly, these ER stress proteins do not co-express with lymphocytes markers such as CD3 and CD20.

Poster 22

Marie Gilon

marie.gilon@student.uliege.be

Laboratory of Human Genetics - Medical oncology
Cancer

Role of eosinophils in clear cell renal cell carcinoma

Gilon M, Boulet D, Thiry J, Chiplunkar S, Waltregny D, Machiels J-P , Bours V, Jerusalem G, Gennigens C, Josse C
Department of Medical Oncology, University Hospital of Liège
Laboratory of Medical Oncology, GIGA Institute, Liège
Department of Urology, University Hospital of Liège
Department of Medical Oncology, Cliniques Universitaires Saint-Luc, Brussels
Department of Human Genetics, GIGA Institute and University Hospital of Liège

Eosinophils (EOS) are mostly known for their role in allergic and anti-parasitic responses. However, recent research has highlighted their role in cancer. Here, we investigate both in vivo and through a retrospective clinical study the impact of EOS in clear-cell renal cell carcinoma (ccRCC). To produce an in vivo model, murine ccRCC cells were implanted into the kidneys of mice. Two groups were established: one receiving injections of anti-IL-5 cytokine to decrease blood EOS count and one control group. A consistent decline in EOS count was observed in the anti-IL5 group compared to the control group ($p<0.0001$), while leukocyte level remained stable over time ($p=0.605$). There was no difference in primary tumor growth between groups when comparing weight ($p=0.438$), but the number of lung metastases was significantly higher in the anti-IL5 group when counted on the lung surface ($p=0.0286$) and on histological sections ($p=0.0099$). The retrospective study involved 182 patients diagnosed with localized ccRCC. Collected data included clinical and anatomopathological characteristics, survival outcomes, hematological values at diagnosis, annually up to five years post-diagnosis, and at progression. As already demonstrated, the neutrophil-to-lymphocyte ratio at diagnosis was correlated with PFS ($p=0.0179$) and OS ($p=0.0211$). A lower relative and absolute EOS counts (respectively REC and AEC) at diagnosis were correlated with a greater risk of disseminated relapse ($p=0.0269$ for REC, $p=0.0233$ for AEC), with a shorter PFS ($p=0.0097$ for REC, $p=0.0304$ for AEC) but there is no association with OS ($p=0.0699$ for REC, $p=0.0549$ for AEC). In conclusion, EOS seem to play a role in metastatic dissemination in our in vivo model, confirmed by the association between EOS count at diagnosis and prognosis for patients diagnosed with localized ccRCC. Experiments must be repeated to confirm those results and investigate further the mechanism by which EOS influence metastatic dissemination.

Poster 23

Florine Habets

florine.habets@uliege.be

Laboratory of Molecular Angiogenesis - Mitochondria in Cell to Cell Communication
Cancer

VEGF-induced Mitochondrial-Derived Vesicles control endosomal maturation in endothelial cells

Habets F, Herkenne S
Université de Liège

Mitochondria, previously seen as static and independent organelles, appear today as major signaling partners within cells, interacting with other organelles and regulating many cellular functions. In our lab, we dove into the study of the regulatory role of mitochondria in angiogenesis, the mechanism by which new blood vessels develop from pre-existing ones. Angiogenesis is central in cancer development as solid tumors rely on the formation of new blood vessels to be supplied with oxygen and nutrients and also to spread via metastasis. It is thus crucial to understand the regulation of this process in order to find new therapies. Angiogenesis involves the activation of endothelial cells and is mainly controlled by the VEGF (Vascular Endothelial Growth Factor) pathway. Although VEGF was primarily seen to favor mitochondrial elongation, we surprisingly noticed the production of mitochondrial-derived vesicles in response to VEGF treatment. These double-membrane vesicles seem to play major role in regulating negatively VEGF pathway by controlling endosomal maturation in endothelial cells. Preventing their production or trafficking leads to an inhibition of endosomal maturation and an overstimulated angiogenesis. Currently, understanding of MDVs biogenesis and trafficking is limited to MDVs produced in oxidative-stress conditions – quality control MDVs. VEGF-induced MDVs seem to be produced by different machineries and have novel regulatory functions in cellular processes. We are studying the biogenesis of VEGF-induced MDVs and the mechanism behind MDV-controlled endosomal maturation.

Poster 24

Robin Jehay

Robin.jehay@uliege.be

Laboratory of Biology of tumor and Development
Cancer

Targeting ferroptosis resistance to enhance sensitivity of cancer cells to radiotherapy

Jehay R, Montero-Ruiz L, Noël A, Foidart P, Sounni NE

Like chemotherapy, radiotherapy is used for the treatment of 50% of human cancers, including lung, brain, breast, head and neck, pancreatic, liver, bone, lung, esophageal, and rectal cancers. Sensitivity to radiotherapy primarily relies on tumor biology, which includes tumor vascularization, expression of DNA damage repair proteins, expression of oxidases, cellular response to reactive oxygen species (ROS), and cellular metabolism. Radiotherapy primarily increases the quantity of ROS via cellular water hydrolysis. This results in accumulation of ROS-induced DNA damage leading to programmed cell death called apoptosis. However, ROS induced by ionizing radiation (IR) can (per)oxidize other molecules such as lipids. Accumulating peroxidized membrane phospholipids triggers a form of cell death known as ferroptosis. Cancer cell resistance to ferroptosis might determine their response to IR. We have defined three breast cancer cell lines with differential response to increasing doses of IR and found that the IR responder cells have increased production of lipide peroxide when compared to cells with intermediate response and resistant cells. Interestingly, cancer cell response to IR was correlated with their expression levels of the stearoyl-CoA desaturase (SCD). The more resistant cells have a higher level of SCD, and the most sensitive cells have a low level of SCD expression. Recently, we demonstrated that mainly lipid desaturation by SCD contribute to tumor recurrence. Here we are exploring the role SCD in conferring resistance to IR and ferroptosis. Furthermore, we are investigating mechanisms driven by lipid desaturation and transport that protect cancer cell from IR and ROS. Our hypothesis suggests that targeting key proteins involved in the lipid metabolism storage and desaturation pathways could enhance the accumulation of lipid peroxide in cell membrane's phospholipids via ROS generated by irradiation, ultimately enhancing IR-induced cell death.

Poster 25

Fanny Lardinois

fanny.lardinois@uliege.be

Metastasis Research Laboratory
Cancer

Blocking glycation stress in chemoresistant colon cancer: Impact on cancer stem cells

Lardinois F, Mohring V, Crake R, Maloujahmoun N, Agirman F, Chariot A, Rapino F, Peulen O, Corbet C and Bellahcène A
Metastasis Research Laboratory, GIGA-Cancer, GIGA Institute, University of Liège
Pole of Pharmacology and Therapeutics (FATH), Institut de Recherche Expérimentale et Clinique (IREC), UCLouvain, Brussels
Cancer Signaling, GIGA Stem Cells, GIGA Institute, University of Liège
Medical Chemistry, GIGA Stem Cells, GIGA Institute, University of Liège

Our team has demonstrated that KRAS-mutated colorectal cancer (CRC) exhibits high methylglyoxal (MG) glycation stress. This stress is linked to glucose metabolism reprogramming during tumor progression and therapy resistance. Evidence suggests that cancer stem cells (CSCs) drive cancer aggressiveness, drug resistance, and tumor relapse. We propose to study MG stress in CRC CSCs.CSCs characterized by high ALDH activity showed significant increases in intracellular MG and MG protein adducts, indicating a correlation between MG and stemness markers. Comparing cells in 2D cultures to 3D spheroids, we found elevated levels of stem cell markers ALDH, OCT-4, and CD133 in 3D cultures. Preliminary experiments revealed increased ALDH1A1 in HT29 spheroids, linked to higher levels of CD44 and cyclin D1, suggesting Wnt pathway activation. RNA sequencing of LIM1215 CRC cells depleted of GLO1 using specific shRNAs showed enhanced expression of Wnt activators, including CTNNB1 (coding for _-catenin), WNT6, WNT7B, WNT9A, LEF1, and TCF4. This suggests that MG stress may contribute to Wnt activation in CRC. Encouraging results show that exogenous MG stress leads to dose-dependent MG adduct accumulation and increased TCF1 expression in HT29 cells. The promotion of stemness by 5FU in CRC suggests that our FOLFOX-resistant models may mimic the sequence of glycolysis, MG stress, and Wnt activation. Neutralizing MG in CSCs could enhance FOLFOX therapy efficacy in CRC. Understanding CSCs' metabolic adaptation to therapy offers promising applications for improving cancer patient care. By exploring these mechanisms, we aim to develop more effective strategies for targeting CSCs and overcoming therapy resistance in CRC, potentially leading to better patient outcomes.

Poster 26

Célia Lemoine

Celia.Lemoine@uliege.be

Laboratory of Nervous System Disorders and Therapy
Neuroscience

Investigation of the cellular and molecular effects of the subventricular zone of the adult brain on hosted glioblastoma cells and the role of these influences in tumor persistence and recurrence

Lemoine C, Dumbruch T, Rogister B, Neirinckx V

Glioblastoma (GBM) is the most common and lethal type of primary brain tumor in adults. It is characterized by high invasiveness and resistance to treatment, leading to systematic relapses. Clinical data and experimental models show that human GBM cells invade the adult brain and find shelter in a neurogenic zone, the subventricular zone (SVZ). Once nested in the SVZ, GBM cells are more resistant to ionizing radiation, suggesting a role of the SVZ in GBM resistance to therapy and relapse. Recently, it was shown that extracellular vesicles (EVs) were important in intercellular communication. EVs play a role in the SVZ and in the GBM. However, the effects of the SVZ-EVs on GBM cells are not known. Our aim is to decipher the effects of the SVZ environment and EVs on SVZ-nested GBM cells. We focus on the neural stem cells (NSCs) populating the SVZ, and on the cerebrospinal fluid (CSF) that is in close contact with this area. Conditioned media (CM) from immortalized human NSCs (hNSC.100) and rat choroid plexus cells (Z310) are used to mimic the NSC secretome and the CSF, respectively. Human CSF samples are also used to confirm the results. EVs are isolated from CM by differential ultracentrifugation. Patient-derived GBM cells are cultured in the different media to assess the morphology, proliferation, invasion and resistance to treatments. We observed that the Z310-CM and the CSF led to an adherent phenotype of GBM cells, normally cultured in suspension. We will also assess the transcriptomic changes of GBM cells cultured in CSF for 24 hours compared to classical medium. In parallel, we grafted patient-derived GBM cells in the brain of immunodeficient mice, dissected SVZin and SVZout GBM, digested the tissues and sorted tumor cells based on RFP expression. RNA was extracted from RFP+ cells from both SVZin and SVZout samples for sequencing and analysis of differentially expressed genes. To conclude, this project will help elucidate the role of the SVZ in GBM progression.
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Poster 27

Chloé Lowette

Chloe.Lowette@uliege.be

Laboratory of Biology of Tumor and development
Cancer

Is there a relationship between uPARAP and the cytoskeleton?

Lowette C, Ivanova E, Deroye J, Pirson S, Gucciardo F, Noël A
Université de Liège, Martin M - Université Libre de Bruxelles

The lymphatic system is involved in lymph drainage and tissue fluid homeostasis, as well as in immune cell transport. Abnormal formation of lymphatic vasculature (lymphangiogenesis) can induce the development of secondary lymphedema following cancer treatment (surgery and/or radiotherapy/chemotherapy). Furthermore, excessive lymphangiogenesis is often associated with metastatic dissemination. We previously assigned an unprecedented and specific role of urokinase plasminogen activator receptor-associated protein (uPARAP, MRC2 gene) in lymphatic endothelial cells (LECs). uPARAP silencing in vitro induced a hyper-permeable and disorganized LEC monolayer. We recently found an unexpected link between uPARAP and the microtubule network. How the cytoskeleton is regulated in LECs is poorly documented. The main objective is here to decipher the interplay between the uPARAP and cytoskeleton organization during cell migration/polymerization and endosomal trafficking occurring during membrane protein endocytosis and recycling.
In our assays, primary human LECs are transfected with a pool of siRNA targeting the MRC2 gene (KD LECs) or a pool of scramble siRNA (CTR LECs). Then, cells are stimulated with VEGFC, the main lymphangiogenic factor. The architecture of the microtubule network and the levels of acetylation and polyglutamylation of tubulin, were similar in KD LECs and in CTR LECs, independently of VEGF-C stimulation. We next analyzed the dynamics of microtubule remodeling through immunostaining of EB1 comet forming on microtubule plus end. Under VEGF-C stimulation, CTR LECs displayed a significant increased number of EB1+ comets as compared with KD LECs. No differences were observed in CTR and KD LECs under basal condition without VEGF-C. Studies are ongoing to investigate how uPARAP could control the dynamics of microtubule plus ends.

Poster 28

Eva Lucarelli

eva.lucarelli@uliege.be

Laboratory of Gene Expression and Cancer
Molecular & Computational Biology

FET fusion oncoproteins rewire alternative splicing patterns to drive sarcomagenesis

Lucarelli E, Ongena L, Dubois L, Galvan B, Bruyr B, Dequiedt F

The FET (FUS, EWSR1, TAF15) genes are commonly involved in chromosomal translocations resulting in their fusion with various transcription factors (TF) genes. These genomic abnormalities are hallmarks of several sarcomas and leukemias, and are found along few other alterations in these neoplasms. The chimeric proteins encoded by these fusion genes share a similar architecture, with a strong aminoterminal transactivation domain derived from FET proteins, and a carboxyterminal DNA-binding domain derived from the TF partner. As this structure is reminiscent of that of a TF, the oncogenic potential of FET fusion proteins has been first attributed to their ability to reprogram transcription. However, this transcriptional role is not sufficient to fully explain how these oncoproteins drive various cancers. Indeed, growing evidence points towards novel post-transcriptional roles for FET fusions, notably in the alternative splicing of pre-mRNA. Such a function has previously been demonstrated for the prototypical FET fusion EWSR1::FLI1, the main driver of Ewing sarcoma.
This project aims to determine whether the pre-mRNA alternative splicing function observed for EWSR1::FLI1 could be a shared mechanism promoting FET fusion-driven oncogenesis. RNA-sequencing of various FET-translocated sarcoma cell lines showed that thousands of alternative splicing events are induced upon FET fusion depletion. In addition, a representative panel of FET fusions promoted exon inclusion of a reporter minigene, but only when directly tethered onto its pre-mRNA, suggesting that the control of splicing by FET fusions could be direct and might rely on its recruitment onto pre-mRNA. The association of FET fusions to RNA was subsequently confirmed in several sarcoma cell lines, and their recruitment might be mediated by intermediary RNA-binding proteins. Finally, we found several splicing events to be controlled by all FET fusions and subsequently assessed the biological relevance of these targets.

Poster 29

Caitlin McIntyre

Cmcintyre@uliege.be

Laboratory of Neuroendocrinology
Neuroscience

The role of AVPV kisspeptin in sexual behaviour

McIntyre C, Evrad F, Beaudou C and Bakker J

Kisspeptin, a key neuropeptide regulating the reproductive axis, has recently been shown to also mediate sexual behaviour in rodents but the underlying neural circuitry remains unknown. In the present study, we utilise a kiss-cre transgenic mouse model to selectively inhibit a hypothalamic population of kisspeptin neurons in the antereoventral periventricular nucleus (AVPV) and determine the effect on sexual behaviour in intact male and gonadoectomised estrogen-replaced female mice. Kiss-cre mice received a viral injection into the AVPV to selectively induce kisspeptin neurons to express either the inhibitory designer receptor exclusively activated by designer drugs (DREADDS), hM4DGi, or as a control, the fluorescent protein mCherry. We found that administration of the chemical actuator Clozapine-N-Oxide (CNO, 2mg/kg) to activate hMD-4Gi and consequently silence AVPV kisspeptin neurons significantly impaired mate partner preference in both male and female mice compared to saline treated and mCherry expressing controls. However, chemogenetic inhibition of AVPV kisspeptin neurons only disrupted sexual behaviour in female mice and had no significant effect in male mice. Collectively, these findings provide further insight into the neural circuitry regulation the expression of sexual behaviours.

Poster 30

Victoria Mohring

victoria.mohring@uliege.be

Metastasis Research Laboratory
Cancer

Impact of Methylglyoxal stress on the immune system within the Breast Cancer Environment

Mohring V , Hubert P, Ancion M, Nokin MJ, Roncarati P, Agirman F, Maloujahmoum N, Lardinois F, Wissocq T, Herfs M, Peulen O, Janji B, Bellahcène A

Metastasis Research Laboratory, GIGA-Cancer, GIGA Institute, University of Liège
Laboratory Experimental Pathology, GIGA-Cancer, GIGA Institute, University of Liège
Tumor Immunotherapy and Microenvironment Group, Department of Cancer Research, Luxembourg Institute of Health (LIH)

The shift to glycolysis in tumor cells has a crucial yet often neglected consequence: the spontaneous production of methylglyoxal (MG). MG is an oncometabolite that can glycate proteins, lipids, and DNA, causing cellular stress. These glycation end products (MG-AGES) promote cancerous traits and support tumor growth and metastasis, as previously shown by our research and others. In the MMTV-PyMT spontaneous breast cancer model, we detected an accumulation of MG-AGES from early adenoma to late carcinoma stages through immunohistochemistry (IHC). We also observed increased recruitment of immunosuppressive granulo-myeloid derived suppressor cells (g-MDSCs) in these tumors, which carnosine, a powerful MG scavenger, effectively inhibited. Similar results were seen in the 4T1 breast cancer syngeneic model. Mice treated with carnosine showed less infiltration of g-MDSCs compared to untreated mice after orthotopic injection of 4T1 breast cancer cells. To further confirm MG stress's role, we generated 4T1 cells with stable depletion of glyoxalase 1 (GLO1), an MG-detoxifying enzyme, thus inducing endogenous MG stress. These MG-stressed breast cancer cells formed primary tumors with high g-MDSC accumulation in their microenvironment, consistent with our findings. Mechanistically, cytokine array analysis under MG stress revealed an increase in cytokines associated with recruiting immunosuppressive cells. Moreover, the efficacy of anti-PD1 antibodies improved when combined with carnosine in the 4T1 grafting model and a cytometry-based multiplex analysis showed that this combination significantly impacted tumor immune infiltration. These results suggest a new link between methylglyoxal and the immune tumor microenvironment, indicating potential therapeutic benefits from combining carnosine treatment with immunotherapy in breast cancer.

Poster 31

Nasrin Mortazavi

nasrin.mortazavi@uliege.be

CRC-Human Imaging
Neuroscience

Locus Coeruleus Activity during Wake Is Associated with Rapid Eye Movement Sleep Intensity

Mortazavi N, Koshmanova E, Sharifpour R, Berger A2, Beckers E, Campbell I, Paparella I, Balda Aizpurua F, Lamalle L, Talwar P, Sherif S, Vandewalle G
Sleep and Chronobiology Lab, GIGA-Institute, CRC-In Vivo Imaging Unit, University of Liège
Institute of Neuroscience (IoNS), Université Catholique de Louvain (UCLouvain), Brussels
Synergia Medical SA, Mont-Saint-Guibert
Faculty of Health, Medicine and Life Sciences, School for Mental Health and Neuroscience, Alzheimer Centre Limburg, Maastricht University; Maastricht, The Netherlands

While the locus coeruleus (LC) appears to be an important structure for sleep in animals, limited imaging studies evaluated whether the LC is related to sleep in humans due to the difficulty of imaging such a small size nucleus. We aimed to investigate the link between LC activity during wakefulness assessed by 7-Tesla MRI and EEG features of sleep. To trigger a response of the LC, we used a visual perceptual rivalry task and an auditory salience detection task. Subjects saw an ambiguous stimulus which could trigger spontaneous switches between two perceptions of the same image. Participants also did an auditory oddball task. 52 healthy volunteers completed the protocol. Participants completed an fMRI session, during which they were administered the two tasks. Participant's sleep was recorded in-lab under EEG to extract 3 sleep features of interest (i.e., rapid eye movement (REM) sleep latency, REM sleep percentage, REM theta energy). We first conducted a general linear model with the SPM12 over the entire brain. We then extracted the activity estimates over individual LC masks and then conducted generalized linear mixed models (GLMMs) to test for associations between the activity of the LC and EEG features of sleep, including age, sex, BMI, and total sleep time as covariates. We identified increased activation within the left LC associated with perceptual switches (uncorrected $p<0.001$; $t > 3.27$) and the detection of the target sound (uncorrected $p<0.001$; $t >3.26$). GLMMs on the LC activity and sleep EEG metrics revealed a positive association between the left LC activity during perceptual rivalry task and REM theta energy ($p = 0.004$, $t=3.02$), which reflects the intensity of REM sleep. A negative association was found between the left LC activity during oddball task and this sleep metric in the older group ($p = 0.024$, $t = -2.33$). These results show that the association between LC activity during wakefulness and REM sleep intensity depends on the cognitive context.

Poster 32

Kim Mottard

kim.mottard@uliege.be

Laboratory of Nervous System Disorders and Therapy
Neuroscience

PTK7 targeting in glioblastoma

Mottard K, Leparc L, Clette B, Vandevenne M, Brans A, Rogister B, Neirinckx V.

Glioblastoma (GBM) is the most aggressive form of primary brain tumor with a median life expectancy of about 16 months after a trimodal therapy. Indeed, due to its high heterogeneity and tissue invasion, this tumor is generally leading to early and fatal relapses after treatment. Analyses of various glioma patient-based datasets showed a high expression of the Protein Tyrosine Kinase 7 (PTK7) in different brain tumors, especially in GBM, compared to non-tumoral brain. PTK7 is a transmembrane protein, firstly discovered as overexpressed in colon carcinoma. This pseudokinase plays a role in development through the modulation of Wnt signalling and has been shown to be involved in cell proliferation, migration, and growth during development and in several cancers. This project therefore aims at investigating the potential of PTK7 as a GBM-specific target for nanobody-based therapeutic approaches. Patient-derived materials obtained from surgical resection allow us to achieve in vitro and in vivo experiments. PTK7 expression was evaluated by immunofluorescence and flow cytometry on patient tissues or patient-derived GBM stem-like cell (GSC) cultures. A PTK7-KO GSC culture was generated based on CRISPR-CAS9. In parallel, anti-PTK7 nanobody sequences were obtained by phage display and their specificity was tested by bio-layer interferometry and flow cytometry analysis. We obtained high expression of PTK7 in GBM samples compared to non-tumoral human brains. Moreover, the expression of this protein is maintained upon ionizing irradiation, temozolomide treatment, and hypoxia. In the future, we will combine this nanobody to the suicide gene therapy induced by oncolytic Adeno-associated virus (AAV), to only target GBM cells. To conclude, PTK7 is overexpressed in GBM tissues and remains stably expressed after therapy-relevant stress conditions in GSCs. This preliminary data supports the potential of PTK7 targeting that could be achieved with high-affinity AAV-nanobodies.

Poster 33

Yanis Mouheb

yanis.mouheb@doct.uliege.be

Sensation and Perception Research Group
Neuroscience

Study of the effects of virtual reality hypnosis as a pain and anxiety relief strategy in oncology patients undergoing a port-catheter placement procedure

Mouheb Y, Vanhaudenhuyse A, Bicego A

The port-catheter (PAC) is a long-term venous access device, frequently used in oncology patients. The PAC placement procedure is an invasive technique, consisting of a reservoir surgically implanted in the chest wall or upper arm, and a catheter in the arm. Patients can experience pain in the incision area after implantation, as well as anxiety before, during and after implantation. Virtual reality hypnosis (VRH) is an innovative technique delivering clinical hypnosis to patients through virtual reality (VR). The clinical applications of VRH are still little known. In this study, we will assess anxiety and pain perception of 102 cancer patients scheduled to receive a PAC. The patients will be randomly divided into two groups. The first group will benefit from the usual care for the placement of the PAC (i.e., local anaesthesia), while the second will benefit from the VRH in addition to the usual care. Pain perception, pain unpleasantness, anxiety, comfort, satisfaction, absorption, dissociation will be evaluated before and after the intervention.

Poster 34

Anna Maria Obeid

annamaria.obeid@uliege.be

Molecular Analysis of Gene Expression (MAGE)
Cancer

Deciphering the role of the putative RNA-Binding Protein TFIP11 in regulating tissue-specific alternative splicing

Obeid AM, Genco A, Galvan B, Flasse L, Pendeville H, Mottet D

RNA-binding proteins (RBPs) comprise a large class of proteins that regulate the metabolism of RNA transcripts throughout their life cycle. Many regulatory RBPs function in a cell-, tissue-, or condition-specific manner and are capable of regulating variety of molecular processes such as alternative splicing of messenger RNAs that is critical for appropriate protein expression in the corresponding tissues. Normal functions of RBPs are vital for human physiology, as defects in RBP function have been associated with many diseases such as neurodegeneration, autoimmune diseases, and cancers. RNA-binding domains (RBDs) in RBPs are the functional units responsible for RNA binding. Multiple RBDs are often present in a single RBP and these modular domains can coordinate and enhance binding to RNA in a sequence and/or structure-specific manner. Tufte-lin-Interacting Protein 11 (TFIP11) is a spliceosome protein which possesses two putative RBDs, one G-patch domain at its N-terminal region and one dsRBD at the C-terminal extremity. Despite we previously demonstrated important regulatory roles for TFIP11 in regulating spliceosome assembly and activation, little is known about the functionality of its RNA-binding modules, nor about the molecular mechanisms governing its role in regulating tissue-specific alternative splicing. The present project aims to further address these two fundamental questions and will provide an unprecedented classification of TFIP11 as a new RNA binding protein.

Poster 35

Morgane Paque

morgane.paque@doct.uliege.be

Laboratory of Cancer Signaling
Cancer

mTORC2-dependent reprogramming of lipid metabolism is a metabolic vulnerability in lung cancer

Paque M, An N, Marques Junior J, Krishnankutty R, Perin F, Leclercq M, Vanleyssem R, Roncarati P, Cataldo D, Berx G, Herfs M, Sumpton D, Le Quesne J, von Kriegsheim A, Close P & Blomme A
Laboratory of Cancer Signaling, GIGA-Institute, University of Liège, Liège
Laboratory of Metabolic Regulation, GIGA-Institute, University of Liège, Liège,
Edinburgh CRUK Centre, Institute of Genetics and Molecular Medicine, University of Edinburgh, Edinburgh, UK.
Laboratory of Tumor and Development Biology, GIGA-Cancer, University of Liège and CHU of Liège
Laboratory of Experimental Pathology, GIGA-Cancer, University of Liège
Cancer Research Institute Ghent (CRIG) and Molecular and Cellular Oncology Laboratory, University of Ghent, Ghent
CRUK Beatson Institute, Garscube Estate, Switchback Road, Glasgow, G61 1BD, UK.
WELBIO department, WEL Research Institute, avenue Pasteur, 6, 1300 Wavre

Lung cancer is a genetically heterogeneous disease characterized by a multitude of tumor-promoting alterations. This heterogeneity renders lung tumors resistant to current treatment strategies. Importantly, genomic amplification of RICTOR, the defining component of mTOR complex 2 (mTORC2), frequently occurs in lung cancer. However, despite high therapeutic potential, targeting mTORC2 activity remains challenging. In this study, we show that elevated mTORC2 signaling in patients with lung adenocarcinoma is associated with poor overall survival and high frequency of TP53 mutation. Furthermore, proteomic characterization of patient biopsies reveal that RICTORhi tumors exhibit a prominent hypoxia phenotype and undergo extensive metabolic rewiring. In order to model this molecular subtype, we have generated a new mouse model of lung cancer by overexpressing Rictor in the lungs of KrasG12D/+-Tp53-/- mice (KPR and KP models respectively). In comparison to the KP model, KPR mice display increased tumor burden and have shortened survival. In line with the patient data, KPR tumors are hypoxic and metabolically reprogrammed towards increased glucose and lipid utilization. Mechanistically, we identified the transcription factor HIF-1_ as a potential mTORC2 target and demonstrate that RICTOR controls HIF-1_ stability through an mTORC2-PKC signaling axis, independently of AKT activity. Using a combination of proteomics, metabolomics and lipidomics, we further demonstrate that HIF-1_ supports lung tumor growth by promoting mTORC2-dependent sphingolipid metabolism. Finally, we show that RICTOR and HIF-1_ are frequently co-expressed in lung cancer biopsies and that this association correlates with poor outcome in lung cancer patients. Taken together, our results support the rationale of targeting mTORC2-dependent lipid metabolism in lung cancer and highlight HIF-1_ as a clinically relevant target for the development of future anticancer therapies.

Poster 36

Léa Pautet

lea.pautet@uliege.be

Molecular Analysis of Gene Expression (MAGE)
Cancer

Deciphering the molecular mechanisms related to proteasome inhibitors resistance in the pathology of multiple myeloma

Pautet L, Besse L, Besse A, Galvan B, Lafontaine DLJ, Mottet D
Molecular Analysis of Gene Expression, GIGA research institute, University of Liege, Liège
Experimental Oncology and Hematology, Department of Oncology and Hematology, Kantonsspital St Gallen, St Gallen, Switzerland
Division of Oncology and Children's research center, University Children's Hospital Zurich, Zurich, Switzerland
RNA Molecular Biology, Fonds de la Recherche Scientifique (F.R.S./FNRS), Université Libre de Bruxelles (ULB), Charleroi-Gosselies

Multiple myeloma (MM) is the second most common blood cancer, with 50000 new cases diagnosed annually in Europe. The median survival of patients is about 5 years, ranging from six months to ten years. Proteasome inhibitors (PI) such as Bortezomib (BTZ) and Carfilzomib (CFZ) are commonly used as first-line therapies. Unfortunately, patients inevitably develop resistance to these treatments. Aberrant RNA splicing is known to be a crucial mechanism in the emergence of cancer resistance to chimiotherapy. To understand the molecular basis of MM resistance, we performed an RNA-seq experiment using BTZ-resistant and CFZ-resistant AMO1 cells compared to sensitive AMO1 cells and analyzed differentially expressed genes as well as altered splicing events.
Among thousands of dysregulated genes, we are interested in downregulation of three Pseudouridine Synthase (PUS) family proteins: PUS1, PUS3 and PUS7. Indeed, PUS proteins are known to isomerize uridine into pseudouridine mainly on tRNAs but also on mRNAs and U snRNAs. Modifications in tRNA pseudouridylation could lead to changes in the translatome of resistant MM cells. Regarding splicing alteration, we found that exon skipping is the main affected event. Interestingly, we found that transcripts encoding for the core proteins - BRE, BRCC36 and MERIT40 - of the two multi-subunit deubiquitination complexes BRISC and BRCA1 are alternatively spliced in PI-resistant cells. These isoforms could alter the assembly and/or function of the BRISC and BRCA1 complexes, thus limiting their abilities to deubiquitinate K63-polyubiquitin chains of their substrates and therefore impacting their functions in key signaling pathways such as DNA repair or inflammatory processes.

Poster 37

John Read

john.read@uliege.be

Imaging - Aging & Memory Lab
Neuroscience

Exploration of the interplay between mental fatigue and effort perception using resting-state functional connectivity

Read J, Guillemin C, Charonitis M, Requier F, Bahri M A, Vandewalle G, & Collette F
GIGA CRC Human Imaging, University of Liège, Liège
Psychology and Cognitive Neuroscience Research Unit, University of Liège, Liège
Department of Neurology, CHU of Liège, Liège
GIGA In Silico Medicine, University of Liège, Liège

The negative impact of mental fatigue (MF) on cognitive performance has long been described but its underlying brain mechanisms remain unclear. It is further postulated that perceived effort may interplay with MF to constitute a stop-signal when the effort-reward is too imbalanced. This study aims to explore the functional connectivity changes in brain regions associated with effort-based decision-making (EBDM) after induction of low or high MF. 17 healthy volunteers (31.42 ± 5.76y.o.) underwent a 2-session fatigue-inducing protocol. MF was induced by manipulating the difficulty of a working memory task with two conditions: slow (control) and fast (MF) items presentation. Participants rated their subjective perception of fatigue and effort pre- and post-task. Following both conditions, participants underwent a 3T resting-state fMRI acquisition. Seed-based connectivity (SBC) analyses were conducted using CONN22.a. Nine seed-regions associated with MF and EBDM were selected a priori. Group-level analyses were thresholded at p<.001 voxel-level and p-FDR<.05 cluster-size (CS). Linear-mixed effect models revealed decline in accuracy with time-on-task in the fatigue condition only (p<.001) and a significant condition*time interaction for the perception of effort (p<.05). We did not find statistical difference regarding the fatigue perception across time and condition. SBC analyses revealed lower functional connectivity in the fatigue compared to control condition for several seeds (all ps-FDR<.05). Among significant results, we observed changes in connectivity: between medial prefrontal cortex (mPFC) and left dorsolateral prefrontal cortex (DLPFC); between bilateral insulas and mPFC. In sum, connectivity is lower in the fatigue condition between key EBDM regions and DLPFC involved in working memory tasks. Importantly, the lower connectivity between inslas and mPFC, respectively thought to be associated with cost-benefit computation, emphasizes the need to take into account the effort needed to perform a task when assessing MF.

Poster 38

Florence Requier

florence.requier@uliege.be

CRC Human Imaging
Neuroscience

Alzheimer's disease is associated with increased cognitive but not physical fatigue: A preliminary study

Requier F, Charonitis M, Lesoinne A, Hostaux Y, Salmon E, Bastin C, & Collette F
GIGA CRC In Vivo Imaging, University of Liège, Liège
Psychology and Cognitive Neuroscience Research Unit, University of Liège, Liège

Fatigue (both physical and mental) is a frequent complaint in neurological disorders and has a deleterious effect on patient's cognition. However, mental fatigue in Alzheimer's disease (AD) has not been investigated, although this population exhibits pathophysiological traits typically associated with pathological fatigue (i.e., structural/functional brain changes, impaired serotoninergic transmission, sleep disturbances…). Therefore, this study aims at investigating the frequency and characteristics of fatigue in AD patients and its relationships with global cognition. 19 patients with mild to moderate AD and 150 healthy controls (HC) completed the Fatigue Scale for Motor and Cognitive Functions (FSMC) measuring physical and cognitive trait fatigue. Global cognitive status was assessed with the MoCA. Linear models were used to determine presence of group effect on cognitive and physical fatigue levels, controlling for age, sex, and education. The two groups differed on age ($p=.04$, AD>HC), sex ($p=.02$, more women in HC) and global cognitive performance ($p<.001$, AD<HC) but not on education ($p=.82$). Higher scores were observed in AD patients for cognitive fatigue (estimate: 5.34, 95% IC: [1.48,9.20], $p=.007$, $R_{sp}=.05$) but not for physical fatigue (estimate: 1.98, 95% IC: [-0.19,6.16, $p=.35$]), with a complaint of cognitive fatigue in 79% of the patients but 44% of HC. Physical fatigue was also reported in 79% of AD patients, and in 63% of HC. No significant association between fatigue level and MoCA performance was observed in AD or HC. These preliminary results indicate that Alzheimer's disease is associated with higher cognitive fatigue, but not physical fatigue. However, the level of cognitive fatigue does not seem related to global cognitive functioning. A better understanding of mental fatigue and its relationships with brain lesions and specific cognitive processes could help to manage this symptom in order to improve cognitive functioning in daily life.

Poster 39

Anne-Sophie Reuter

as.reuter@uliege.be

Unit of Animal Genomics
Molecular & Computational Biology

Viral clonal landscape and tumor progression: lessons from tumor-resistant sheep in the BLV leukemia model

Reuter A-S, Wayet J, Shahzad N, Griebel P, Arsic N, Facciuolo A, Garigliany M-M, Vroomen S, Burny A, Georges M and Van den Broeke A
Unit of Animal Genomics, GIGA, ULiège, Liège
Laboratory of Viral Oncogenesis, Institut Jules Bordet, ULB, Brussels
VIDO, University of Saskatchewan, Saskatoon, Canada
Département de morphologie et pathologie, ULiège, Liège

Bovine Leukemia Virus (BLV), a deltaretrovirus closely related to Human T-cell Leukemia Virus-1 (HTLV-1), induces B-cell leukemia/lymphoma in ~5 % of infected individuals following several years of asymptomatic infection. Early stages of infection are characterized by a large number of clones, each uniquely identified by their proviral integration site in the host genome until one of these clones suddenly expands, leading to cancer. Experimentally infected sheep systematically develop B-cell leukemia after a shorter asymptomatic period, characterized by a gradual increase in proviral load (PVL), providing a unique in vivo model for studying tumor progression. Here, we have the opportunity to screen an unusual cohort of sheep infected with miRNA-impaired BLV variants which we found resistant to tumor development. We generated a bio-bank of longitudinal samples from wild-type and variant BLV-infected sheep (>11 years, > 100 time-points), providing an unlimited source of genetic material covering each stage of the disease, and investigated their insertional landscape using a viral clonality NGS method developed by our team. Unlike WT-infected sheep, animals infected with miRNA-impaired variants were characterized by low and stable PVLs, and none of them developed malignancy. The clonal landscape of these animals is also drastically different, as indicated by the loss of a strong hotspot signature, the absence of a significant orientation bias of closely-located proviruses and their scattered distribution, although proviral insertion sites remain highly enriched in cancer driver genes. The tumor-resistant cohort showed a mono/oligoclonal-like distribution with long-term surviving dominant clones while such signatures were not observed in WT-infected asymptomatic animals at any PVL. Using this cohort and single-cell NGS technologies, we characterized the viral reservoir of infected B-cells and their microenvironment to identify transcriptional changes underlying this unusual long-term asymptomatic phenotype and its unique clonal landscape. Studying features from clonal architecture to transcriptome signatures in long-term surviving sheep will contribute to our understanding of the mechanisms underlying tumor development.

Poster 40

Anaïs Servais

anais.servais@uliege.be

CRC Human Imaging
Neuroscience

Exploring a novel virtual environment benefits memory performance: Effect of exploring a novel environment or rather of a new experience?

Servais A, Schomaker J, Bastin C.
GIGA Research, CRC Human Imaging, University of Liège
Faculty of Social Sciences, Leiden University, the Netherlands

Whether exposure to a novel environment in virtual reality improves subsequent verbal memory or not is still debated. The present study is an attempt to replicate previous positive findings. To do so, we assessed whether exploration of a novel virtual environment leads to better word memory than exploration of a familiar environment and whether this effect is related to exploratory behavior. Fifty-five young participants completed three sessions. The first session consisted of assessing the baseline memory performance. In this memory task, participants encoded a list of 40 words and then freely recalled as many words as possible, immediately and after a 24-hour delay. In the second session, participants started with a familiarization phase with a virtual environment and then explored the familiar or a novel environment before performing a memory task. In the third and last session, participants explored either the familiar or novel virtual environment (according to the condition they did not fulfill in the second session) before performing the memory task. Like some other prior studies, our results failed to find a positive impact of exploring a novel environment (i.e., spatial novelty) on memory performance. Rather, we observed that memory recall is higher after exploring a virtual environment (regardless of it being novel or familiar) compared to the baseline performance. In fact, the experience of virtual reality might induce another type of novelty, called distinct novelty, which could impact memory too. We discuss how future studies could try disentangling the effect of these two types of novelty on subsequent memory performance. We believe this is a necessary step to understand the conflicting results between existing studies.

Poster 41

Fatemeh Seyfzadehdarabad

f.seyfzadehdarabad@uliege.be

Coma Science Group
Neuroscience

Paradoxical Responses to Zolpidem in the General Population

Seyfzadehdarabad F, Mat B, Sanz LRD, Martial C, van der Lande G, Lejeune N, Thibaut A, Hanson J, Boly M, Laureys S, Gosseries O

Zolpidem is a non-benzodiazepine hypnotic agent commonly prescribed for sleep disorders. Initially administered to manage restlessness in severe brain-injured patients with disorders of consciousness (DoC), it paradoxically elicits consciousness recovery in _5-20% of these patients. Similar paradoxical effects (e.g., inability to fall/stay asleep, higher levels of concentration, agitation) are observed in the general population, yet the prevalence rate is unknown. This cross-sectional epidemiological study aims to (1) investigate the prevalence of zolpidem use and its paradoxical effects in the general population, (2) determine onset time and duration of these effects, and (3) identify common paradoxical effects. Hence, an anonymous online survey was distributed. Participants were recruited through (1) network (social media and institutional websites) and (2) crowdsourcing approaches. The inclusion criteria were ≥18yo and fluency in English and/or French. Excel and R software were used for data cleaning and analyses. Out of 15,289 participants (mean age 32 ± 13; 52% female; from 84 countries), 4% (n=785) reported zolpidem use and 16% (n=128) reported paradoxical effects. Paradoxical effects most frequently emerged within the first hour (58%) and lasted up to 6 hours (60%). Inability to fall asleep (60%), restlessness (42%), inability to stay asleep (35%), excitation (35%), agitation (26%), increased alertness (19%), increased energy (19%), increased concentration (10%), and other (11%) (e.g., euphoria, anxiety) were reported as paradoxical effects. Binary logistic regression analyses showed a history of neurological or psychiatric disorders could significantly predict paradoxical responses to zolpidem (CI: 1.08 – 2.56; $p<0.05$). Yet, potential mechanisms underlying zolpidem effects (paradoxical vs. non-paradoxical) are still unclear. Therefore, studies with multimodal approaches are warranted to investigate these mechanisms in DoC patients and the general population.

Poster 42

Marlyne Squatrito

Marlyne.Squatrito@uliege.be

Laboratory of Biology of Tumor and Development
Cancer

From Pathway to Patient – Transforming Adenomyosis Treatment with S1P Inhibitors

Squatrito M, Vervier J, Bindels J, Bernet L, Miceli S, Nisolle M, Munaut C.

Adenomyosis is a benign uterine disorder characterized by the infiltration of endometrial glands and stroma within the myometrium. Previous studies, including our own, have identified significant immune system alterations in this condition, such as abnormal distributions of immune cell populations in the uterus in both human and murine models. The Sphingosine-1-Phosphate (S1P) pathway, critical in regulating immune cell trafficking, cell proliferation and vascular function, is suspected to play a role in these immune disturbances. Our study aimed to explore and investigate new potential therapeutic treatments by investigating how the S1P pathway is modulated in the uteri of mice with adenomyosis and examining the effects of fingolimod, an S1P receptor modulator, on disease progression in both in vitro and in vivo settings. Adenomyosis was induced in CD1 mice through tamoxifen gavage from the first day of life for four consecutive days. We compared the expression of S1P pathway components in uteri from mice at 1 month and 3 months of age. Adenomyosis-induced mice were treated daily with fingolimod starting at six weeks of age for three weeks. Post-treatment, the mice were sacrificed, and their uteri were analyzed to assess lesion size and immune cell modulation. FACS analysis evaluated the impact of fingolimod on immune cell populations. Additionally, the effect of S1P on cell proliferation was examined in vitro using human endometrial stromal cells (hESCs). We observed modulation in the expression of S1P receptors and the enzymes responsible for S1P synthesis in the uteri of mice with adenomyosis. While fingolimod treatment did not completely restore immune disturbances, it significantly reduced lesion size. In vitro, S1P stimulated hESC proliferation. These findings highlight the potential of targeting the S1P pathway as a therapeutic approach in adenomyosis. Future research will focus on testing other S1P inhibitors and exploring different timings of their administration to assess their potential in preventing lesion formation, cell proliferation, cell migration and immune regulation.

Poster 43

Marty Stepniak

mstepniak@uliege.be

Laboratory of Translationnal Gastroenterology
Immunobiology

Study of the role of the epithelium in the initiation and progression of intestinal fibrosis in Ulcerative Colitis

Stepniak M, Adam P, Salée C, Loly J-P, Vieujean S, Kropp S, Reenaers C, Vankemseke C, Coimbra C, Decker E, Massot C, Bletard N, Delvenne P, Speca S, Louis E & Meuwis M-A.

The mechanisms of initiation and progression of intestinal fibrosis in ulcerative colitis (UC) are poorly understood. No antifibrotic medical treatment, nor marker exists to highlight intestinal fibrosis in inflammatory bowel disease and fibrosis increases patient complication rate and decrease quality of life. ER stress (ERS) is present in the epithelial cells of UC tissues with inflammation and fibrosis. Our team demonstrated that the protein AGR2 was increased in epithelial cell lines under ERS and that extracellular AGR2 (eAGR2) released in media was able to induce intestinal fibroblast to myofibroblast transition (FMT). We have developed a translational model capable of modeling the involvement of the epithelium to study its impact in the development of fibrosis in the context of ulcerative colitis (UC): the apical-out organoids derived from UC intestinal crypts. These organoids (n=6) shows that after the application of transient ER stress induced by tunicamycin, a relative expression of AGR2 which is increased between 2 and 19 fold compared to the vehicule condition (Tris). Other ER stress markers (BIP, CHOP, sXBP1) also persist during the post-stimulation period with fold changes ranging between 8 to 19, 5 to 10, and 4 to 8, respectively. These organoids also shows a pro-fibrotic paracrine effect on an intestinal fibroblast cell line by inducing a fibroblast-to-myofibroblast transition (FMT) detected by an _-SMA increase of up to 3 in certain conditions compared to control. AGR2 signal on stimulated organoids shows an increased and more diffuse cytoplasmic localization compared to the vehicule conditions where it is highly visible at the membrane. Therefore, we present a fibrogenic intestinal organoids model capable of modeling the response of the UC epithelium after ERS. This model will be used to clarify the role of the epithelium and its pro-fibrotic paracrine action (and associated molecular changes) in intestinal UC fibrosis

Poster 44

Puneet Talwar

ptalwar@uliege.be

Sleep and Chronobiology Unit
Neuroscience

Age-related changes in the association between REM sleep and the polygenic risk for Parkinson's disease

Talwar P, Mortazavi N, Koshmanova E, Collette F, Maquet P, Phillips C, Vandewalle G
GIGA-CRC Human Imaging, Université de Liège

Parkinson's disease (PD) is one of the rare diseases for which sleep alteration is a true marker of disease outcome. How the association between sleep and PD emerges over the healthy lifetime trajectory is not known. We examined association between polygenic risk score (PRS) for PD and the variability in the sleep electrophysiology in a large sample of 415 individuals. In this prospective observational study, in-lab EEG recordings of habitual sleep were conducted with automatic scoring of sleep stages, artefact detection and additional energy bands computation. We also extracted DNA from saliva/blood for PRS determination. Summary statistics of large PD GWAS was used to compute PRS using SBayesR approach. Generalized Additive Model for Location, Scale and Shape (GAMLSS) regression showed significant association for REM duration and theta in REM as dependent variables with PD PRS while controlling for age, sex, BMI and total sleep time or REM duration. In the younger cohort, the analysis revealed positive association of REM duration (p=.002; _:0.004) and REM theta energy (p =.019; _: 0.01) with PD PRS. In contrast the analysis of the older cohort, revealed negative association of REM duration (p=.004; _:-0.01) with PRS and yielded a significant interaction of age with PRS for REM theta energy (p<.0001; _: 0.003). Our findings show that REM sleep is associated with the polygenic risk for developing PD in age dependent manner. A higher risk for PD is associated with higher REM duration and REM sleep intensity during early adulthood while it is associated with lower REM duration and REM sleep intensity after 50y. This reveals a switch in the association between younger, presumably free of alpha-synuclein inclusions, and older healthy individuals in which low levels of these inclusions may be present. These findings may contribute to unravelling the core association between PD and sleep and to the identification of novel intervention targets to prevent or delay PD.

Poster 45

Thomas Trevisan

thomas.trevisan@uliege.be

Laboratory of Tumor and Development Biology
Cancer

MICA shedding by the TF/FVIIa complex: impact on tumor cell resistance to NK cell cytotoxicity

Trevisan T, Goïta A, Villard A, Gilles C

Tissue Factor (TF) is a membrane protein overexpressed by certain cancer cells and has emerged as a central player linking coagulation and cancer. TF is mainly known as the primary initiator of the coagulation cascade, serving as the transmembrane receptor for the first proteolytic factor of the coagulation cascade, FVIIa. Adding to coagulation-dependent pro-metastatic functions documented by us and many others, TF has also been shown to promote tumor progression through hemostasis-independent mechanisms such as the activation of the Protease-Activated Receptor 2 (PAR2), inducing its signaling and promoting tumor growth and angiogenesis. At the basis of this project, MHC Class I polypeptide-related sequence A (MICA) was identified as a substrate of the TF/FVIIa proteolytic complex, opening the possibility that TF/FVIIa could modulate Natural Killer (NK) cells/tumor cells interactions. MICA is a membrane protein expressed by cells undergoing cellular stress, such malignantly transformed cells, and is the one of the major activating ligands of the NKG2D receptor expressed by NK cells, which leads to their activation in the circulatory system. A well-known mechanism of immune evasion employed by tumor cells is the proteolytic shedding of MICA by canonical sheddases of the ADAM/MMP families. Our team is exploring whether TF/FVIIA-mediated MICA shedding is a similar yet distinct mechanism to ADAM/MMP shedding, being likely activated in different body locations. Western blot analysis of breast tumor cell line MDA-231and lung tumor cell line Calu1 conditioned media showed a TF/FVIIa-dependent release of a ~30 kDa soluble MICA fragment, distinct from the ~50 kDa fragment released by canonical sheddases. Additionally, FACS analysis showed that FVIIa treatment on MDA231 cells induced a depletion of MICA at the membrane level. In vitro cytotoxicity co-culture assays are ongoing to determine the relevance of this depletion regarding NK cell cytotoxic function modulation.

Poster 46

Vincent Wuidar

vincent.wuidar@uliege.be

Laboratory of Biology of Tumor and Development
Cancer

Prevention of breast cancer growth: a complex interplay between estrogen receptor activation and estrogen nature

Wuidar V, Dias Da Silva , Zielonka M, Flas C, Staumont B, Pequeux C

Menopausal hormone treatment (MHT) based on estrogen preparation increases breast cancer risk and development, mainly by stimulating the proliferation of estrogen receptor (ER_)-positive cells. We hypothesized that the nature of the estrogen and its route of administration could prevent this pro-tumorous effect of MHT. Indeed, distinct pharmacokinetic profiles are associated to specific routes of estrogen administration. In contrast to oral treatment reaching a steady-state, pulsed estrogen therapy is characterized by early high and transient peak of estrogen plasma concentration. Despite similar efficacy to oral MHT, the impact of pulsed estrogen therapy on breast cancer and ER_ activity is still unknown. Using human breast cancer cell lines, patient-derived xenograft (PDX), protein and transcriptomic analysis, we compared the impact of three natural estrogens, estradiol (E2), estriol (E3) and estetrol (E4) on ER activation and breast cancer growth, when they are administered either continuously or in a pulsed manner. We observed that, in contrast to E2, pulsed E3 and E4 treatments only transiently activate ER_ signaling, preventing breast cancer growth. However, continuous estrogen (E2, E3, E4) treatments increase breast cancer growth through continuous ER_ signaling activation. This study emphasized that the estrogen nature and its route of administration both impact breast cancer progression.

Poster 47

Can Yuan

can.yuan@uliege.be`

Unit of Animal Genomics
Molecular & Computational Biology

Evaluation of heritability partitioning approaches in livestock populations

Yuan C, Duarte Gualdron JL, Takeda H, Georges M and Druet T

Heritability partitioning approaches estimate the contribution of different functional classes, such as coding or regulatory variants, to the genetic variance. This information allows a better understanding of the genetic architecture of complex traits, including complex diseases, but can also help improve the accuracy of genomic selection in livestock species. However, methods have mainly been tested on human genomic data, whereas livestock populations have specific characteristics, such as high levels of relatedness, small effective population size or long-range levels of linkage disequilibrium. Here, we used data from 14,762 cows, imputed at the whole-genome sequence level for 11,537,240 variants, to simulate traits in a typical livestock population and evaluate the accuracy of two state-of-the-art heritability partitioning methods, GREML and a Bayesian mixture model. In simulations where a single functional class had increased contribution to heritability, we observed that the estimators were unbiased but had low precision. When causal variants were enriched in variants with low (<0.05) or high (> 0.20) minor allele frequency or low (below 1st quartile) or high (above 3rd quartile) linkage disequilibrium scores, it was necessary to partition the genetic variance into multiple classes defined on the basis of allele frequencies or LD scores to obtain unbiased results. When multiple functional classes had variable contributions to heritability, estimators showed higher levels of variation and confounding between certain categories was observed. In addition, estimators from small categories were particularly imprecise. However, the estimates and their ranking were still informative about the contribution of the classes. We also demonstrated that using methods that estimate the contribution of a single category at a time, a commonly used approach, results in an overestimation. Finally, we applied the methods to phenotypes for muscular development and height and estimated that, on average, variants in open chromatin regions had a higher contribution to the genetic variance (> 45%), while variants in coding regions had the strongest individual effects (> 25-fold enrichment on average). Conversely, variants in intergenic or intronic regions showed lower levels of enrichment (0.2 and 0.6-fold on average, respectively). Heritability partitioning approaches should be used cautiously in livestock populations, in particular for small categories. Two-component approaches that fit only one functional category at a time lead to biased estimators and should not be used.

Poster 48

Manon Zielonka

Manon.Zielonka@uliege.be

Laboratory of Biology of Tumors and Development
Cancer

Could pulsed estrogen-based therapies enhance the sensitivity of breast cancer to endocrine therapies and prevent resistance ?

Zielonka M, Wuidar V, Dias Da Silva I, Schyns W, Jerusalem G, Péqueux C
Laboratory of Biology, Tumors and Development (LBTD), Estrogen-sensitive tissues team, GIGA-cancer, University of Liège
Clinical oncology Department, GIGA-cancer, University of Liège

While endocrine therapies have been successful in reducing estrogen receptor (ER)-positive breast cancer recurrence and mortality in early-stage patients, about 20% still face recurrence due to resistance to these therapies in metastatic disease. It is crucial to identify new strategies to prevent endocrine resistance. Our team generated preliminary results showing that pulsed estrogen treatments increase the expression of ER in vitro and in vivo in endocrine sensitive breast cancer cells. We hypothesized that a pulsed administration of estrogen could increase the sensitivity of ER-positive breast cancer to endocrine therapy and thus could prevent or overcome resistance. Our aim was to characterize ER expression in parental and resistant cells after estrogen pulsed treatments and to evaluate the efficacy of endocrine therapies. Our results show an increase of ER expression not only in parental cells, but also in several resistant cells to Palbociclib and Fulvestrant treated with pulsed administrations of estradiol. However, pulsed treatments are not able to prevent the loss of ER expression in late stages of MMTV-PyMT tumors. Interestingly, we observed an increase of sensitivity of parental cells to tamoxifen. Then we evaluated the impact of Elacestrant, a new endocrine therapy, on ER expression. In conclusion, pulsed administration of estrogen increases the expression of ER by parental and resistant cancer cells and could sensitize cells to endocrine therapies. The next goal is to study the effect of pulsed treatment on the sensitivity of resistant cells to endocrine therapies by in vitro and in vivo experiments.

Poster 49

Mikhail Zubkov

m.zubkov@uliege.be

Sleep and Chronobiology Laboratory
Neuroscience

R2*, R1, and proton density age variations in subcortical structures in young adults at 7T

Zubkov M, Pine K, Bazin PL, Mortazavi N, Talwar P, Lamalle L, Phillips C, Collette F, Alkemade A, Kirilina E, Weiskopf N, Vandewalle G.

Ultra-high field (UHF) MRI and its ability to provide high resolution data is progressively entering the clinical application stage and provides promising advances for brain imaging. Quantitative MRI (qMRI) is a promising technique in UHF MRI, aiming to remove the protocol- and scanner-dependency in MRI by measuring parameters related to the biophysical properties of the tissues such as relaxation rates R1, R2, and R2*, proton density, magnetization transfer saturation and others. Here, we present a preliminary evaluation of variation of a number of qMRI parameters with age within a sample of young and healthy volunteers for a multiparameter mapping (MPM) qMRI protocol. A cohort of healthy subjects underwent the scanning at 7T Terra MRI. The resulting images were processed with the open source hMRI toolbox, yielding R2*, PD and R1 maps. Subcortical structures were automatically parcellated by Multi-contrast Anatomical Subcortical Structures Parcellation (MASSP) using all three parametric maps as input. Median values for each of the parameters were extracted for the subcortical structures. A linear model was built for each of the median value sets. We present the linear models with p-values for age lower than 0.05 (false discovery rate-corrected).

Poster 50

Benedetta Zuliani

benedetta.zuliani@uliege.be

Laboratory of Gene Expression and cancer
Molecular & Computational Biology

The role of m6A RNA modification during EndMT

Zuliani B, Ongena L, Claes M, Bruyr J, Dequiedt F

In response to environmental and mechanical stimuli, fully differentiated endothelial cells (ECs) can dedifferentiate into mesenchymal cells in a process termed endothelial-to-mesenchymal transition (EndMT). The tumor microenvironnement is characterized by a proinflammatory context, hemodynamic abnormalities and hypoxia, all of which are known to promote EndMT. Recently, several studies have suggested that EndMT might be controlled by epigenetic mechanisms. On this note, N6-methyladenosine (m6A), the most common mRNA internal modification, has recently gained attention in the field of epitranscriptomics through its link with differentiation processes. However, the precise role of m6A modifications during EndMT has not been investigated yet. Our project aims to: 1) investigate the role of the m6A epitranscriptomic machinery in EndMT, 2) identify transcripts whose m6A content is specifically affected during EndMT, and 3) test the hypothesis that specific transcription factors (TFs) might be involved in defining the EndMT-associated m6A epitranscriptome. We established by RT-qPCR, western blotting and phalloidin staining that the absence of METTL3 and METTL14, members of the complex responsible for m6A deposition on RNA, slow down the EndMT induced by IL-1_ and TGF-_2 in ECs, demonstrating the role of the m6A machinery in the promotion of the EndMT. Furthermore, we performed m6A-seq2 and highlighted thousands of transcripts whose m6A content varies during the EndMT, a significant portion of which include TFs. We decided to focus on a m6A deposition on the 3'UTR of SNAI1, a TF involved in the early steps of EndMT, and we demonstrated that differential methylation affects expression and stability of the transcript. Our results provide a mechanism of regulation of cancer progression by modulation of EndMT through RNA m6A modification.

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