

10th Annual Conference Aspects of Neuroscience

Abstract Book

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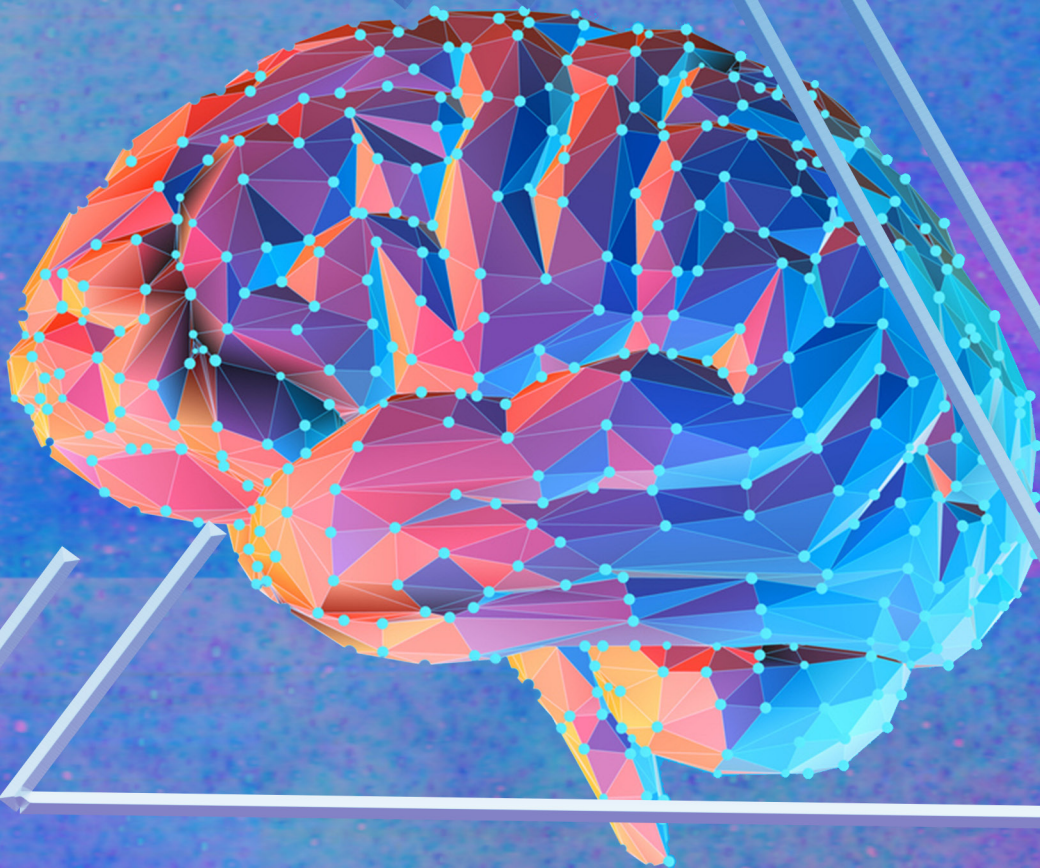
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26-28<sup>th</sup>  
November  
2021



**X<sup>th</sup> Aspects of  
Neuroscience**

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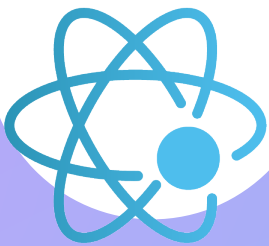
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Dear Colleagues,

Welcome to the 10th International Conference Aspects of Neuroscience!

We, Aspects of Neuroscience Organizing Committee, are very glad that gathering brilliant neuroenthusiasts during the last weekend of November at the Faculty of Physics, University of Warsaw, became our tradition and one of the keypoints among Polish neuroscientific events. Aspects of Neuroscience is also a brand, recognized by members of scientific institutions from Poland and many more European countries. It is our privilege to host both experienced researchers and students from all over the globe who come to Warsaw in order to present their research, receive feedback from their peers and get mutually inspired.

The programme condenses, unequivocally, the dynamic, challenges and relevant contribution of neuroscience research in the broader context of human health and pushing the boundaries of knowledge even further. Our aim is to give you opportunity to listen to some of the leading researchers of different aspects of neuroscience: Biological, Computational, Cognitive and Clinical. We firmly believe that cooperation above disciplines and integration of neuroscientists will result in new ideas and thinking outside the box.



# **X INTERNATIONAL CONFERENCE “ASPECTS OF NEUROSCIENCE”**

Three days of the Conference are literally packed with events of high meritorical value: seven plenary lectures of internationally recognized professors, dozens of very interesting short speeches given by PhD and Msc students and around sixty posters. The level of presented seminar speeches and posters is maintained with the help of the Scientific Committee. Each abstract submitted had at least a double peerblinded review. Moreover, we encouraged our experts to participate in a panel discussion “Finding hope in science”. However, the Conference offers you even more than fantastic scientific experiences. We put much energy to bring participants closer together and to show you the beauty and power of our great region. Furthermore, we encourage you to join us for the Integration Party on Saturday, when you can forget for a moment about the neuroscientific buzz and relax with your colleagues (and perhaps become more familiar with delicious drinks). We hope that you will have a wonderful and engaging time at this Conference. Members of our Organizing Team will be available at any time if you have a question or need any help.

Warm regards from the AoN21 Team!



The conference is organized by the members of Neuroinformatics Scientific Student Association at the University of Warsaw and by student volunteers from many Polish universities.

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Marta Paż

Head of the VII International Conference “Aspects of Neuroscience” Krzysztof Bielski

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# **EVENT PROGRAM DAY BY DAY**

**26 NOVEMBER 2021 (FRIDAY)**

10:30 Registration desk open!

18:00 – 18:15 Opening Ceremony

18:15 – 19:15 Opening Lecture (online)

Prof. Lars Kai Hansen – “Machine learning for brain state monitoring based on EEG”



# EVENT PROGRAM DAY BY DAY 27

## NOVEMBER 2021 (SATURDAY)

09:00 – 10:00 Plenary Lecture (on-site)

Prof. Michał Żochowski – “Understanding neural dynamics underlying memory consolidation during sleep”

10:10 – 11:50 Seminar Session: Neurobiological

- Gabriela Izowit – “Brain state-dependent responses of midbrain dopaminergic neurons to the aversive stimulus”
- Dominik Kanigowski – “Learning-related changes in intrinsic excitability of GABAergic interneurons in the somatosensory cortex of mice”
- Aleksandra Mielnicka – “Do neurons modulate exocytotic gliotransmission in astrocytes?”
- Mateusz Kostecki – “The influence of the socially acquired information on the brain representation of space”

11:50 – 12:20 Coffee break

12:20 – 13:20 Plenary Lecture (on-site)

Prof. Peter Achermann – “Cortical neuronal activity and sleep homeostasis”

13:30 – 14:20 Seminar Session: Clinical

- Jan Szczypiński – “Abnormal behavioral and neural responses in the right dorsolateral prefrontal cortex, during emotional interference on cognitive control in pedophilic sex offenders”
- Agata Szlaga – “Ventral tegmental area-originating dopaminergic modulation of nucleus incertus to interpeduncular nucleus input – possible neuronal mechanism in control of novelty preference expression”

14:20 – 15:30 Lunch break

15:30 – 17:00 Experimental Poster Session: Neurobiological & Clinical, Theoretical Poster Session

17:00 – 18:00 Plenary Lecture (on-site)

Prof Hartwig R. Siebner – “A brain-circuit perspective for the treatment of brain disorders”

18:00 – 19:00 Discussion panel “Finding hope in science”

# EVENT PROGRAM DAY BY DAY

28 NOVEMBER 2021 (SUNDAY)

09:00 – 10:00 Plenary Lecture (online)

Prof. Karl Friston – “Me and my Markov blanket”

10:10 – 11:25 Seminar Session: Cognitive

- Agnieszka Adamczyk – “Threat perception and regulation: Investigating the effect of evolutionary relevance”
- Agata Wolna – “How second language use impacts speech production in the native language: an fMRI study using functional localizers”
- Łucja Doradzińska – “Awareness, attention, and threats - exploring the limits of unconscious fear reaction”

11:25 – 12:00 Coffee break

12:00 – 13:00 Plenary Lecture (on-site)

Dr Agnieszka Grabska-Barwińska – “How to train deep networks in the brain? A fresh take on ‘deep learning’ inspired by the cerebellum”

13:10 – 14:00 Seminar Session: Computational

- Wiktor Rorot – “Modeling “spatial purport of perceptual experience”: egocentric space perception in a virtual environment”
- Piotr Biegański – “A physicist’s view on the analysis of actigraphic bedtime data”

14:00 – 15:00 Lunch break

15:00 – 16:30 Experimental Poster Session: Computational & Cognitive

16:30 – 17:30 Plenary Lecture (online)

Prof. Rafał Bogacz – “Computational models of reinforcement learning”

17:35 Award Ceremony and Closing remarks

# KEYNOTE SPEAKERS





# PhD Agnieszka Grabska-Barwińska

Agnieszka Grabska-Barwińska is a Research Scientist at Google DeepMind, London. She owns a Masters in Physics (MISMaP, Warsaw University) and a Ph.D. in Computational Neuroscience (IGSN, Ruhr University Bochum). She received additional training in Experimental Neuroscience (with Dirk Jancke, RUB) and Machine Learning and Bayesian approaches to the brain (with Peter Latham, University College London). In DeepMind, she swapped the goal of understanding intelligence in vivo for understanding intelligence in silico. This was meant to be easier! She is still immersed in analysing Artificial Intelligence.

## *How to train deep networks in the brain? A fresh take on “deep learning” inspired by the cerebellum*

The dominant view in neuroscience is that changes in synaptic weights underlie learning. It is unclear, however, how the brain is able to determine which synapses should change, and by how much. This uncertainty stands in sharp contrast to deep learning, where changes in weights are explicitly engineered to optimize performance. However, the main tool for doing that, backpropagation, is not biologically plausible, and networks trained with this rule tend to forget old tasks when learning new ones. Here we introduce the Dendritic Gated Network (DGN), a variant of the Gated Linear Network (Veness et al., 2017), which offers a biologically plausible alternative to backpropagation. DGNs combine dendritic “gating” (whereby interneurons target dendrites to shape neuronal response) with local learning rules to yield provably efficient performance. They are significantly more data efficient than conventional artificial networks and are highly resistant to forgetting, and we show that they perform well on a variety of tasks, in some cases better than backpropagation. The DGN bears similarities to the cerebellum, where there is evidence for shaping of Purkinje cell responses by interneurons. It also makes several experimental predictions, one of which we validate with in vivo cerebellar imaging of mice performing a motor task.

# Professor Karl Friston

Karl Friston is a theoretical neuroscientist and authority on brain imaging. He invented statistical parametric mapping (SPM), voxel-based morphometry (VBM) and dynamic causal modelling (DCM). These contributions were motivated by schizophrenia research and theoretical studies of value-learning, formulated as the dysconnection hypothesis of schizophrenia. Mathematical contributions include variational Laplacian procedures and generalized filtering for hierarchical Bayesian model inversion. Friston currently works on models of functional integration in the human brain and the principles that underlie neuronal interactions. His main contribution to theoretical neurobiology is a free-energy principle for action and perception (active inference). Friston received the first Young Investigators Award in Human Brain Mapping (1996) and was elected a Fellow of the Academy of Medical Sciences (1999). In 2000 he was President of the international Organization of Human Brain Mapping. In 2003 he was awarded the Minerva Golden Brain Award and was elected a Fellow of the Royal Society in 2006. In 2008 he received a Medal, College de France and an Honorary Doctorate from the University of York in 2011. He became of Fellow of the Royal Society of Biology in 2012, received the Weldon Memorial prize and Medal in 2013 for contributions to mathematical biology and was elected as a member of EMBO (excellence in the life sciences) in 2014 and the Academia Europaea in (2015). He was the 2016 recipient of the Charles Branch Award for unparalleled breakthroughs in Brain Research and the Glass Brain Award, a lifetime achievement award in the field of human brain mapping. He holds Honorary Doctorates from the University of Zurich and Radboud University.

## *Me and my Markov blanket*

This presentation offers a heuristic proof (and simulations of a primordial soup) suggesting that life—or biological self-organization—is an inevitable and emergent property of any random dynamical system that possesses a Markov blanket. This conclusion is based on the following arguments: if a system can be differentiated from its external milieu, then the system's internal and external states must be conditionally independent. These independencies induce a Markov blanket that separates internal and external states. Crucially, this equips internal states with an information geometry, pertaining to probabilistic beliefs about something; namely external states. This free energy is the same quantity that is optimized in Bayesian inference and machine learning (where it is known as an evidence lower bound). In short, internal states will appear to model—and act on—their world to preserve their integrity. This leads to a Bayesian mechanics, which can be neatly summarised as self-evidencing. I will try to unpack these ideas using simulations and relate them to predictive processing and sentient behaviour.

# Professor Rafał Bogacz

Rafał Bogacz graduated in computer science at Wrocław University of Technology in Poland. Then he did a PhD in computational neuroscience at the University of Bristol, and next he worked as a postdoctoral researcher at Princeton University, USA, jointly in the Departments of Applied Mathematics and Psychology. In 2004 he came back to Bristol where he worked as a Lecturer and then a Reader. He moved to the University of Oxford in 2013. His research is in the area of computational neuroscience, which seeks to develop mathematical models describing computations in the brain giving rise to our mental abilities. He is particularly interested in modelling the brain networks involved in action selection and decision making, and understanding how brain dynamics change in Parkinson's disease.

## *Computational models of reinforcement learning*

A great advance in understanding brain networks underlying reinforcement learning has been achieved thanks to joint contributions of experimental and computational neuroscience. The synergy between these fields started with an observation that dopaminergic neurons in the brain encode the same reward prediction error signal that was used in reinforcement learning algorithms from artificial intelligence. Since then, computational models were developed to describe how this signal is generated and how it modulates learning in brain areas innervated by dopaminergic projections. This talk will start with an overview of classical models describing how the neural circuits in the basal ganglia learn about expected rewards. Then it will discuss more recent models of how the basal ganglia also learn about reward uncertainty, and their relationship to experimental data.

# Professor Hartwig R. Siebner

Hartwig Roman Siebner is heading the Danish Research Centre for Magnetic Resonance (DRCMR) at Copenhagen University Hospital Hvidovre and is full professor for Precision Medicine at the Faculty of Health and Medical Sciences, University of Copenhagen. He is a board-certified neurologist and has previously worked as researcher at Technische Universität München (1995-2000), University College London (2000-2002), and Christian Albrechts University Kiel (2002-2008). Hartwig Roman Siebner combines functional and structural brain imaging with non-invasive transcranial brain stimulation (NTBS), pharmacological challenges, or training interventions. He uses this combined intervention-mapping approach to infer causal dynamics in functional brain networks and to gain insights into brain plasticity and the pathophysiology of brain diseases (movement disorders, multiple sclerosis). His current research is geared to transform NTBS into a precision therapy for personalized treatment of brain circuit disorders.

## *A brain-circuit perspective for the treatment of brain disorders.*

Most, if not all, brain diseases are “circuit disorders” in which aberrant neural circuit activity determines individual functional impairment. Transcranial brain stimulation (TBS) techniques have attracted increasing interest given their unique potential for manipulating dysfunctional brain circuits directly. Yet, circuit targeting is often imprecise and not geared to the individual brain-circuit dysfunction. In my talk, I will make the following points: (i) Leveraging insights from basic and human systems neuroscience will render it possible to transform TBS into a precise tool for personalized brain-circuit modulation. (ii) Focusing on Parkinson’s disease and major depression as prototypical circuit disorders, I will show how in-depth brain mapping of aberrant circuit activity can identify individual brain-circuit dysfunctions. (iii) I will argue that personalized TBS regimes are needed to target the identified dysfunctions by optimizing extrinsic (dosing) and intrinsic (state) variables of TBS. (iv) Optimization requires online and offline brain mapping to probe target engagement and modulation. The insights yielded by brain mapping will allow to refine the personalized TBS approach based on the TBS-evoked circuit changes and clinical improvement.

# Professor Lars Kai Hansen

Professor Lars Kai Hansen is with the Technical University of Denmark, where he heads DTU Compute's Section for Cognitive Systems. His research concerns machine learning with applications in bio-medicine, digital media and cognitive systems. He has published more than 300 papers on these subjects in journals, conferences, and books. The major contributions include introduction and analysis of neural network ensembles (1990-96); the first application of predictive machine learning for mind reading in PET (1994-) and fMRI (1997-); the concept of cognitive components (2005-); the cure for variance inflation (2001, 2011-). Recent work concerns real-time attention monitoring using EEG. His research is generously supported by Danish and international research councils and foundations, including the US National Institutes of Health, the European Union and the Danish Lundbeck and Novo Nordisk Foundations. He served as Cátedra de Excelencia at Universidad Carlos III de Madrid and is a fellow of the European Laboratory for Learning and Intelligent Systems (ELLIS).

## *Machine learning for brain state monitoring based on EEG*

Electroencephalography (EEG) is a widely used non-invasive technology for brain state monitoring. The EEG signal is complex and confounded by artifacts, hence modelling and decision making based on EEG is a serious challenge. I will review progress in machine learning for EEG analysis and applications including attention monitoring, sleep scoring, microstate inference for biomarkers in computational psychiatry.

# Professor Michał Żochowski

Professor Michał Żochowski is affiliated with the Department of Physics and The Biophysics Program at University of Michigan. He and his research group focus mainly on dynamical aspects of information processing in the brain from the perspective of pattern formation in complex networks. To understand those mechanisms, they connect theoretical as well as experimental approaches. Theoretical studies focus on synchronization and dynamical control in simple nonlinear systems, as well as in more complex, biologically feasible, computational models. They are especially interested in coupled systems with self-adaptive units that could model neuromodulatory processes in the neural systems. From the experimental side, they employ optical imaging systems (CCD camera and/or photodiode array) to monitor activity of large neuronal populations.

## *Understanding neural dynamics underlying memory consolidation during sleep*

Why do we sleep? Why after sleep we often remember rehearsed information or routine better than before? It was discovered about a century ago that sleep facilitates memory consolidation however mechanisms underlying that phenomenon are still not known. At the same time it is widely accepted the network dynamics underlying given brain function, is a complex outcome of network structure, modulatory states and incoming external input. In this talk I will focus on neuromodulatory effects of acetylcholine (ACh) and the role it can play in sleep dependent memory consolidation. I will build the modeling framework starting from understanding how ACh affects the excitation properties of individual neurons, then continue to show how these properties modulate network dynamics and finally hypothesize how network-wide dynamics mediate reorganization of sleep representations that lead, via STDP, to memory consolidation. The model predictions are closely compared to analysis of experimental data on sleep dependent contextual fear memory consolidation in mice.

# Professor Peter Achermann

Peter Achermann is Emeritus Professor at the University of Zurich, where he previously led a research group focused on basic and clinical aspects of sleep regulation and analysis of electroencephalography (EEG) signals. He also served as scientific director of The KEY Institute for Brain-Mind Research in Zurich until March 2021. Prof. Achermann is internationally recognized for his contributions to sleep EEG analysis, modeling of sleep regulation and circadian rhythms, and investigations related to biological effects of electromagnetic fields. His reputation as a top sleep EEG expert, in particular in relation to EEG analysis, is reflected in numerous international collaborations and scientific publications throughout his career. Over the past decade, Prof. Achermann's research has mainly focused on trait aspects in sleep and wake EEG, developmental aspect of sleep and sleep regulation in preschool children and adolescents, brain connectivity and cerebral blood flow during sleep, and the process of sleep onset and the borderland between wakefulness and sleep. Recent methodological advances included machine learning, particularly deep learning, to automatically score sleep stages and detect microsleep episodes (short fragments of sleep < 15 s).

## *Cortical neuronal activity and sleep homeostasis*

According to traditional theory, the need for sleep accumulates during wakefulness and dissipates during sleep. A key notion is that sleep-wake history determines the levels of homeostatic sleep pressure, referred to as Process S which is reflected in electroencephalogram (EEG) slow wave activity (SWA, EEG power in 0.5–4 Hz range) during sleep. It has been widely used to obtain insights into sleep regulatory mechanisms. The notion of sleep as a local, activity-dependent process suggests that neuronal activity history must be integrated to determine the dynamics of global Process S. We developed novel mathematical models of Process S based on cortical activity recorded in freely behaving mice, describing the local Process S as a function of the deviation of neuronal firing rates from a locally defined set-point, independent of the global sleep-wake state. Averaging locally derived Processes S and their rate parameters yielded values resembling those obtained from EEG SWA and global vigilance states. Although time awake is considered to be the main variable affecting sleep need, investigating the role of additional extrinsic influences on the dynamics of Process S remains essential to understand its neurophysiological substrates. Surprisingly, a combination of experimental and modeling approaches to investigate the influence of waking behavior and time of day on sleep homeostasis revealed that the mechanisms underlying Process S dynamics are mostly resilient to external factors.

# **SPEECHES**





# CLINICAL SESSION

Szczypiński J.1,2\*, Wypych M.2, Krasowska A.1, Wiśniewski P.1, Kopera M.1, Suszek H. 3, Marchewka A.2, Jakubczyk A.1, Wojnar M.1,4\*

1Department of Psychiatry, Medical University of Warsaw, Warsaw, Poland;

2Laboratory of Brain Imaging, Nencki Institute of Experimental Biology of Polish Academy of Sciences, Warsaw, Poland;

3Faculty of Psychology, University of Warsaw, Warsaw, Poland;

4Department of Psychiatry, University of Michigan, Ann Arbor, MI, USA;

## ***Abnormal behavioral and neural responses in the right dorsolateral prefrontal cortex, during emotional interference on cognitive control in pedophilic sex offenders***

Research from the last decade show neural and behavioral differences between men with pedophilic disorder who commit child sexual abuse (CSA+), and those who do not commit such offences (CSA-).

We aimed to study differences in brain function and behavior, between CSA+ and CSA-, related to emotional interference of cognitive processes.

We recruited three groups of participants CSA+ (n = 11); CSA- (n = 15) and matched healthy controls (HC; n = 17). Participants performed an emotional Go/NoGo in a block-design fMRI experiment. The task comprised four conditions: Negative Go - including only Go stimuli and negatively valenced pictures; Negative NoGo - including 50% of Go and 50% of NoGo trials and negatively valenced pictures; and two corresponding conditions with neutral pictures. The brain analysis was restricted to right and left dorsolateral prefrontal cortex (DLPFC) using small volume correction.

We found a significant interaction of group and valence on reaction times. Direct comparison showed that HC and CSA- had significantly slower reactions in negative compared to neutral blocks, which was not observed in CSA+ group. On the brain level, we found a significant group difference related to the effect of emotional interference (Negative > Neutral), in the right DLPFC but not in the left. Namely, CSA+ showed lower activity than HC and CSA-.

In CSA+ negative distractors failed to increase cognitive control processes, which was observed in CSA- and CSA+ on behavioral and neural level, in line with previous studies showing that cognitive and emotional impairments are linked to offender status rather than pedophilic interests.

# CLINICAL SESSION

Agata Szlaga, Patryk Sambak, Anna Blasiak

Department of Neurophysiology and Chronobiology, Institute of Zoology and Biomedical Research, Jagiellonian University in Krakow

## ***Ventral tegmental area-originating dopaminergic modulation of nucleus incertus to interpeduncular nucleus input – possible neuronal mechanism in control of novelty preference expression***

Proper discrimination between novel and familiar stimuli and adequate responses to novelty are crucial not only for proper functioning but also survival. Importantly, many neuropsychiatric disorders (such as anxiety, autism, schizophrenia and attention deficit hyperactivity disorders (ADHD)) manifest in atypical reaction to novelty. One of the predisposing factor for the development of novelty response-related deficiencies is stress. In this regard, we got interested in nucleus incertus (NI) to interpeduncular nucleus (IPN) innervation. NI is a highly stress-sensitive brainstem structure also involved in arousal, locomotion and memory formation. NI is reciprocally connected with the IPN, located in the midbrain, which was shown to play a role in novelty/familiarity recognition and behavioural inhibition. At the same time, neurotransmitter dopamine (DA) has an established role in motivation-related processes and preference expression, and it was shown that ventral tegmental area (VTA) dopaminergic neurons increase their activity upon exposure to novel stimuli, which then lead to dopamine release.

The aim of our studies was to investigate the functional connectivity between NI and IPN, as well as possible interactions of NI originating innervation and dopaminergic transmission, via D1 receptor, at the level of single IPN neuron. In order to test whether IPN neurons converge NI originating signals known to be involved in stress and arousal control, with novelty related DA/D1R signalling, whole-cell patch-clamp recordings of IPN neurons activity were combined with optogenetic stimulation of NI- originating fibres and bath application of D1R agonist (SKF81297, 10uM). Moreover, to unravel the source of DA in the IPN, viral vector-based retrograde tract-tracing experiments combined with immunofluorescent staining against tyrosine hydroxylase (TH, marker of monoaminergic neurons) were performed.

Upon optogenetic stimulation of NI-originating fibres mostly inhibitory light-evoked postsynaptic currents (le IPSCs) were observed in the IPN. Moreover, 63% of recorded IPN neurons exhibited increase in inward current after D1R agonist application, suggesting that they belong to 'novelty pathway'. Among recorded neurons sensitive to optogenetic stimulation of NI-originating fibres, 61.5% was activated by D1R agonist, showing that the same IPN neurons are sensitive to both novelty-related dopamine signalling and stress-related signals from the brainstem. Notably, analysis of the shape of le IPSC revealed, that D1R agonist application led to decrease in their amplitude. Tract-tracing experiments showed that VTA is the main source of DA in the IPN. Interestingly, results of these experiments also revealed that non-dopaminergic, TH negative, presumably GABAergic, VTA neurons also innervate IPN. Strikingly, whereas vast majority of TH positive, retrogradely labelled VTA neurons were localized near the midline, TH negative IPN-innervating neurons were localized in lateral parts of the structure, suggesting that VTA→IPN innervation signals both salience (DA signalling) and possibly value (nonDA) This electrophysiological and anatomical data suggests a possible role of nucleus incertus – interpeduncular nucleus – ventral tegmental area neuronal loop in the control of novelty preference and attentional processes.

# COGNITIVE SESSION

Agata Wolna<sup>1</sup>, Jakub Szewczyk<sup>2</sup>, Michele Diaz<sup>3</sup>, Aleksandra Domagalik<sup>4</sup>, Marcin Szwed<sup>1</sup>, Zofia Wodniecka<sup>1</sup>

1 Institute of Psychology, Jagiellonian University, Kraków, Poland

2 Donders Institute for Brain Cognition and Behaviour, Radboud University, Nijmegen

3 Social, Life, and Engineering Sciences Imaging Center, the Pennsylvania State University, Pennsylvania, United States

4 Malopolska Centre of Biotechnology (MCB), Jagiellonian University in Krakow, Krakow, Poland

## ***How second language use impacts speech production in the native language: an fMRI study using functional localizers.***

When bilingual speakers switch back to speaking in their native language (L1) after having used the second language (L2) even for a short time, they often experience a difficulty in retrieving words in their L1: a phenomenon referred to as the L2 after-effect. Most likely, the L2 after-effect reflects the increased difficulty in accessing words in L1 in consequence of control processes engaged during prior L2 use (e.g. Guo et al., 2011; Branzi et al., 2014; Wodniecka et al., 2020; Branzi et al., 2016). However, it is unclear whether the L2 after-effect reflects increased demands imposed on the general cognitive control system or effects confined to the language network.

In the present study we address this question by looking at the neural basis of the L2 after-effect in an fMRI experiment using a series of localizer tasks allowing us to identify subject-specific language networks (Fedorenko et al. 2010) and Multiple Demand networks (Duncan et al., 2010). Forty-one Polish-English bilinguals named pictures in L1 and L2. Each participant completed two sessions: in one of them the critical block of naming pictures in L1 was preceded by a short exposure to L1 (naming in L1 after L1) and in the other one, by a short exposure to L2 (naming in L1 after L2). Next, we compared mean BOLD activations between the two conditions in group-constrained subject-specific ROIs defined based on the localizer tasks. We found that the increase in brain activity related to speaking in L1 after L2 was limited to ROIs within the multiple demand network (bilateral anterior frontal pole, right MFG and left STG) with no differences within the language-specific ROIs.

Overall, our results suggest that the L2 after-effect is driven by domain-general control mechanisms and not by changes in activation within the language network. This implies that the L2 after-effect is a result of engagement of domain general mechanisms rather than a passive decrease of the activation level of L1 language representations.

# COGNITIVE SESSION

**Agnieszka K. Adamczyk**

Psychophysiology Lab, Institute of Psychology, Jagiellonian University

## ***Threat perception and regulation: Investigating the effect of evolutionary relevance***

Because humans are biologically prepared to fear evolutionary relevant threats (e.g., snakes and spiders), these types of threats constitute the most common objects of specific phobias. Phobic responses are strong emotional reactions towards phobic objects, which can be described as a deficits in the automatic emotion regulation (ER). Importantly, difficulties in the voluntary control of fear to such stimuli in phobics suggest that emotional responses to evolutionary versus modern threats can be also less prone to effortful ER. Thus, in the present study, we investigated whether emotional responses to evolutionary threats can be differently subject to cognitive ER than modern threat also in healthy, young, and non-phobic adults. EEG signal was recorded from 61 female participants while they reappraised, distracted from, or passively watched pictures of attacking snakes (evolutionary threat, ET) or aimed handguns (modern threat, MT). We measured Early Posterior Negativity (EPN, 225-300 ms), a parieto-occipital component that reflects early automatic attention, and centro-parietal Late Positive Potential (LPP, early: 450-1000 ms, and late time-window: 1000-3000ms), an electrocortical marker of sustained motivated attention and emotional arousal, which served as an index of ER effects. Results revealed that although ET versus MT received facilitated attentional processing at both early (EPN) and late (early LPP) processing stages, responses to both ET and MT were successfully downregulated with the use of both ER strategies. However, comparing each strategy to the passively watch condition revealed differences in timing and magnitude of the observed ER effects: while distraction modulated attention from as early as 450 ms after picture onset (early and late LPP time-window), reappraisal did so only after 1s (late LPP). Moreover, between-strategy comparison demonstrated that distraction was more effective than reappraisal in down-regulating not only objective (the LPP), but also subjective (negative experience) indices of emotional responding to either type of threat. Our findings show that despite differences in early and automatic processing of ET versus MT, fear responses evoked by both types of threats are equally susceptible to ER via effortful and voluntary cognitive ER; however, distraction might show superiority over reappraisal in regulating immediate fear responses, regardless of the (evolutionary or modern) origin of the threat that evoked them.

# COGNITIVE SESSION

**Łucja Doradzińska, Michał Bola**

Laboratory of Brain Imaging, Nencki Institute of Experimental Biology, Polish Academy of Sciences, Warsaw, Poland

## *Awareness, attention, and threats - exploring the limits of unconscious fear reaction*

The ability to quickly and effectively react in the face of danger is crucial for survival in a complex, unpredictable environment. Therefore it has been proposed that the coordinated response to threatening events might begin already at the unconscious stages of perception (LeDoux, 1998). Indeed a compelling body of evidence indicates that subliminal signals of threat can induce fear reaction and enhance unspecific physiological arousal (Tamietto & de Gelder 2010). However, it remains unclear whether an unconscious fear can engage cognitive processes such as attention (Hedger et al., 2016). Thus in the present study we aimed to investigate the power of invisible signals of threat to capture selective attention and direct cognitive resources.

In the conducted experiment we presented participants (N = 41) with pairs of faces. Faces were located on the opposite sides of the screen and each of them could express either fear or a neutral emotional state. Faces were displayed for 16 ms and followed either by an empty screen (supraliminal, conscious condition) or a backward mask, which interfered with visual processing resulting in subliminal perception (unconscious condition). Each trial ended with the presentation of two dots located on one of the sides of the screen. The experiment was divided into two tasks: a dot-probe task and face identification task. In the former participants were asked to respond to the arrangement of two dots (vertical or horizontal), thus faces were treated as task-irrelevant distractors. In the latter participants had to identify emotional expression of a face followed by the dots, therefore faces were considered as task-relevant targets. In order to measure the evolution of brain response related to detection and attentional prioritization of face stimuli we recorded the EEG signal and extracted event related potentials (ERP).

ERP analysis revealed that early, perceptual P100 component reacted similarly to fearful and neutral faces in all consciousness and task conditions. The following, face specific N170 ERP potential was greater when two fearful faces were presented in comparison to two neutral faces. This effect was present independently from stimulus awareness and ongoing task. In contrast, the N2pc component - which indicated that attention was shifted to a fearful face when it was paired with a neutral face - was observed only in the conscious condition. The ongoing task showed no influence on N2pc amplitude. Finally, task-evoked prioritization of fearful faces, reflected by increased amplitude of P300 component, was present only when stimuli were perceived supraliminally.

Our results demonstrate that signals of threat can be detected without awareness and that this happens automatically, irrespective of the task performed. However, although subliminal signals of threat were detected, we found no evidence that they attracted attention unconsciously. According to our findings both automatic, and task-related attentional selection of threatening stimuli can occur only on conscious stages of perception. Our conclusions add to the discussion on subliminal origin of fear suggesting that specific, targeted reaction to threats requires conscious processing.

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# COMPUTATIONAL SESSION

**Piotr Biegański**

Faculty of Physics, University of Warsaw, Warsaw, Poland

## *A physicist's view on the analysis of actigraphic bedtime data*

Actigraphy is a simple and noninvasive method of monitoring human activity, based upon data recorded from accelerometers, embedded usually in watch-like devices. It is widely used in the field of sleep quality assessment, where it has proven to be cheaper and more convenient, yet not largely inferior, alternative to polysomnography (PSG). While the recording devices are nowadays simple and cheap, major challenges appear in the field of analysis and interpretation of recorded signals.

Over the course of almost 40 years many algorithms have been developed in order to discriminate the actigraphic data gathered during bedtime to sleep and wake. Such binary time series may be then used to construct various indices allowing to assess e.g. sleep quality.

Algorithms investigated in this presentation are: Cole-Kripke algorithm, Sazonov algorithm, Webster algorithm, Scripps algorithm, and the UCSD algorithm. All of them were derived empirically by fitting coefficients of a given equation so that the concordance with scored PSG data is maximal. However, the formulae itself were chosen in most cases ad hoc, without proper argumentation. After processing the signal with the obtained formula, the output was thresholded, and each point was classified wake or sleep depending on crossing the threshold.

In our research we propose a mathematical framework fully describing the action of these algorithms. It appears that all these algorithms may be broken down into the following three stages: non-standard resampling to very low frequency (1/60 or 1/30 Hz), application of low-pass filter with cutoff frequency corresponding to period of about 15 minutes, and optional application of rescaling rules, which aim to smoothen the scored signal a bit in a nonlinear way. It seems that all these stages contribute to smoothing the input signal, with an idea that thresholding the smoothed signal should give classification into wake and sleep.

Analysis of these algorithms in terms of the underlying mathematics /signal processing reveals some potential shortcomings, as for example methods used in the first stage (downsampling) may introduce artifacts to the signal due to aliasing. On the other hand, some limitations listed by the authors in original papers seem to not be really problematic—e.g. some aspects of how the signal is downsampled seem to play a marginal role, contrary to algorithm authors' intuition.

We also aim to further test our approach by collecting data from parallel recordings (PSG with actigraphy) gathered during bedtime. Next important step towards systemization of this field is to create a package for actigraphic data analysis and processing, which we are currently working on, with the aim to provide robust and shared as an open-source, versatile software, hopefully paving a way towards clarification and unification of processing and analysis of the actigraphic data.

# COMPUTATIONAL SESSION

**Wiktor Rorot**

University of Warsaw, Warsaw, Poland

## ***Modeling “spatial purport of perceptual experience”: egocentric space perception in a virtual environment***

Egocentric space perception is a multimodal skill, closely tied to action and bodily movements and has an inherent phenomenal dimension. While allocentric space representation has attracted wide attention among psychologists and neuroscientists and resulted in a fairly good understanding of key neural mechanisms, egocentric space is still less well known.

One prominent account, “Skill Theory” provided by Rick Grush (2007), has focused on posterior parietal cortex (PPC) as a key neural area. In Grush’s proposal, the phenomenal experience of spatiality, called “spatial purport of perceptual experience”, has been underlied by embodied, anticipatory mechanisms, with central role assigned to multimodal interfacing between various sensory modalities, as well as bodily skills. Grush has proposed a computational model based on a Kalman filter for the operation of relevant areas of PPC, to account for the appearance of “spatial purport”. Although this model is compatible with dominant accounts of PPC and existing neuroscientific evidence, hitherto it has not been empirically verified directly in relation to egocentric space representation.

The talk reviews Grush’s model and presents a study providing a direct computer simulation of this model in a biologically inspired virtual environment. The goal of the simulation was to develop an agent with a realistic ability for egocentric space perception and representation based on biologically plausible neural network approximations of Kalman filter and Grush’s Skill Theory. To achieve this goal, we use machine learning techniques, with a strong focus on unsupervised and reinforcement learning methods. Resulting agent is tested behaviorally on ecologically plausible tasks to evaluate its internal, learned representations. While this is still a work in progress, initial simulation results indicate that some elements of Grush’s proposal, particularly formalizing the emulation system in terms of a Kalman filter, are implausible.

Finally, the talk will discuss how this type of behaviorally-oriented computer simulation can be used to study phenomenal experience and its qualities, pointing out assumptions, limitations and promises of employed methods.



# NEUROBIOLOGICAL SESSION

Izowit G.<sup>1</sup>, Walczak M.<sup>1</sup>, Drwięga, G.<sup>1</sup>, Pradel, K.<sup>1</sup>, Solecki, W.<sup>2</sup>, Błasiak T.<sup>1</sup>

<sup>1</sup>Department of Neurophysiology and Chronobiology, Institute of Zoology and Biomedical Research, Jagiellonian University, Cracow, Poland <sup>2</sup>Department of Neurobiology and Neuropsychology, Institute of Applied Psychology, Jagiellonian University, Cracow, Poland

## *Brain state-dependent responses of midbrain dopaminergic neurons to the aversive stimulus*

Ventral tegmental area (VTA) and substantia nigra pars compacta (SNc) located in the midbrain constitute the main source of dopamine in the mammalian brain. Dopaminergic (DA) neurons of VTA and SNc have been repeatedly shown to generate burst of action potentials in response to a reward or cue associated with it, and cease to fire in response to reward omission or aversive stimulus. For years it has been assumed that, in this manner, midbrain DA neurons uniformly encode the information about value of perceived stimuli. This notion was revised by the identification of DA neurons that are excited by both rewarding and aversive stimuli, regardless of their value. This resulted in division of midbrain DA neurons into two distinct subpopulations: one encoding the value and the other encoding salience of perceived stimuli. It was also shown that the general state of the brain modulates electrical activity of the midbrain DA neurons, but it remains unknown whether this factor may also influence signalling of the value and/or salience. Therefore, we wanted to determine whether DA neurons can encode information about value and salience of perceived stimuli in brain state-dependent manner. For this purpose, we combined optogenetic tagging and extracellular in vivo recordings of VTA and SNc DA neurons' responses to the electrical footshock applied to the urethane anaesthetised rats. Besides previously described value- and salience-coding neurons we have also observed subpopulation of DA neurons that was not discovered so far. There is a relatively large subpopulation of DA neurons that are inhibited by footshocks during REM-like brain state but with the appearance of non-REM-like phase, these neurons change their response to excitation. It can be hypothesised that this subpopulation may be involved in 'dual-coding' of both value and salience of stimulus depending on the general state of the brain. Recognizing that ongoing brain state determines how certain DA neurons respond to stimuli may provide new scientific ideas about how information about value and salience can be processed.

# NEUROBIOLOGICAL SESSION

**Aleksandra Mielnicka**

Nencki Institute of Experimental Biology, Polish Academy of Sciences, Warsaw, Poland

## *Do neurons modulate exocytotic gliotransmission in astrocytes?*

Over two decades ago, the interplay between neuronal connections and astrocytic processes was described in the tripartite synapse model. It assumes that neurotransmitters activate metabotropic receptors on astrocytes leading to increases in cytosolic  $\text{Ca}^{2+}$  levels via release from endoplasmic reticulum (ER), which then causes vesicular release of gliotransmitters. Numerous controversies arose over the years regarding this model, therefore in our work we try to address some of this criticism and take a closer look at regulated astrocytic exocytosis. We use Synaptobrevin2 (vesicle-associated membrane protein) in fusion with pHluorine (pH-dependent GFP) to observe gliotransmitters secretion in primary mixed hippocampal cultures as well as in pure astrocytic cultures. Our results indicate that the rate of vesicular gliotransmission in astrocytes is higher in pure astrocytic cultures than in mix cultures of neurons and glia, where blocking neuronal transmission with TTX does not significantly reduce the rate of exocytosis. However, we have shown that electrical stimulation of mix hippocampal culture increases the rate of gliotransmitters release even when  $\text{Ca}^{2+}$  release from ER is blocked by 2-APB and Ryanodine. Additionally, activation of group I metabotropic glutamate receptors with DHPG in pure astrocytic and in mix culture increases the rate of exocytosis, indicating a role of  $\text{Ca}^{2+}$  released from ER. On the other hand, we have shown that incubating cells with  $\text{Ca}^{2+}$  chelator - BAPTA decreases the frequency of vesicles released by astrocytes in both types of cultures proving importance of extracellular  $\text{Ca}^{2+}$  in gliotransmitters exocytosis. To conclude our results indicate that there is a link between neuronal component, neuronal activity,  $\text{Ca}^{2+}$  concentrations, and the rate of gliotransmission process in astrocytes. This study is supported by the National Science Centre, Poland grant 2017/26/D/N23/01017.

# NEUROBIOLOGICAL SESSION

**Dominik Kanigowski, Joanna Urban-Ciećko**

Laboratory of Electrophysiology, Nencki Institute of Experimental Biology, Warsaw, Poland

## ***Learning-related changes in intrinsic excitability of GABAergic interneurons in the somatosensory cortex of mice.***

The cortex consists of two types of neurons that cooperate to maintain the function of the brain: (1) excitatory (glutamatergic) neurons and (2) inhibitory (GABAergic) interneurons. Excitatory neurons account for more than 80% of neurons in the cerebral cortex. Nonetheless, local GABAergic inhibitory interneurons play an integral role in the appropriate functioning of the central nervous system. Their population is very diverse and consists of cells with various biochemical, anatomical, and electrophysiological features. This heterogenic group of cells is classified based on the expression of three main protein markers: somatostatin (SST), parvalbumin (PV), and ionotropic serotonin 5HT<sub>3a</sub> receptor. The last class is further split into vasoactive intestinal polypeptide- (VIP) and non-VIP-expressing interneurons. The vast diversity of GABAergic interneurons allows them to cope with multi tasks which they are responsible for in the brain. Many literature pieces of evidence show that GABAergic interneurons are responsible for memory formation, learning, and plasticity. In our laboratory, we explore learning-related plastic changes of GABAergic interneurons at the functional level. In our experiments, we employ a simple model of conditioning in mice where two types of stimuli are paired in time. Tactile conditioned whisker stimulation is linked with unconditioned electric tail shocks. Subsequently, whole-cell patch-clamp recordings from acute cortical slices are done to look for plastic changes in electrophysiological properties of GABAergic interneurons in the primary somatosensory cortex after animal's conditioning. We found an increase in intrinsic excitability of low-threshold spiking SST-expressing cells in layer 4 in the cortical representation of stimulated whiskers in a conditioned group of animals. Next, we observed that pseudoconditioning leads to a decrease of intrinsic excitability in layer 4 fast-spiking PV-expressing interneurons. In contrast, layer 4 VIP interneurons characterize higher complexity in response to learning. The plasticity of VIP cells varies in the case of their electrophysiological type and form of learning.

Intrinsic excitability of accommodating VIP (but not low-threshold spiking) cells increases in conditioned animals in relation to the pseudoconditioned group. The results above show that alterations in intrinsic excitability are related to all analyzed types of GABAergic interneurons, however, learning-related plastic changes of the GABAergic system are characteristic for specific types of interneurons and forms of learning. Therefore plastic changes in the inhibitory system can be indispensable for right memory formation and learning. Funding: National Science Centre, Poland UMO-2015/18/E/NZ4/00721 to J.U.C.

# NEUROBIOLOGICAL SESSION

**Mateusz Kostecki**, Martyna Pałys, Julia Bochniarz

Laboratory of Emotions Neurobiology, Nencki Institute of Experimental Biology, PAS, Warsaw, Poland

## *The influence of the socially acquired information on the brain representation of space*

Animals gather information about the environment from many sources. The most obvious one is exploration - they can learn about the localization of food, mates or predators by exploring the environment by themselves. On the other hand, animals living in groups can acquire this information from conspecifics - and this information can guide their further exploration of space. In my project, I study how mice transfer information about a distant food source; I have found that they can extract this information during a short interaction with another mouse. Moreover, this socially acquired information changes the motivational value of explored space, as indicated by an increased activity in olfactory tubercle, a region associated with estimation of a motivational value of odour, during an exploration of the environment. Moreover, I test the changes in the brain representation of space in CA1 region of hippocampus associated with socially acquired information about space.



# POSTER SESSION



# CLINICAL SESSION

**Alicja Szadziewska**

## *Non-invasive diagnosis of Alzheimer's disease - new approaches*

Alzheimer's disease becomes more prevalent as the population ages. Robust and reproducible biomarkers for early diagnosis are essential for research and clinical practice, as well as clinical trials. Core cerebrospinal fluid biomarkers (A $\beta$ 42, T-tau and P-tau) are not useful for routine patient assessment due to invasive collection of the specimen. Research groups currently give priority on development of diagnostic techniques utilising blood or saliva as tested biological samples. They require minimally invasive procedures and are more cost-effective than PET imaging.

This review discusses the newly developed blood tests for the detection of amyloid- $\beta$  and p-tau, neurofilament light and glial fibrillary acidic protein and their potential for extensive application in clinical medicine. Saliva as a source of diagnostic biomarkers is currently explored. It's metabolomic and proteomic analysis display changes that are associated with Alzheimer's disease. Neurofilament light and lactoferrin were proposed as saliva biomarkers.

Experiments investigating Alzheimer's disease biomarkers for prediction, differential diagnosis, prognosis and progression assessment could result in new standard of patient care.



# CLINICAL SESSION

**Izabela Chałatkiewicz, Marta Agnieszczak, Hanna Górecka, and Monika Tutaj**

## *Does N400 reflect the preserved consciousness? On language processing in disorders of consciousness patients.*

Disorder of consciousness (DoC) is a state of prolonged altered consciousness and includes coma, unresponsive wakefulness syndrome (UWS) and minimal consciousness state (MCS). UWS patients are thought to be unaware of themselves and their surroundings, in turn, MCS patients show some traces of preserved consciousness. Nevertheless, the accurate diagnosis of the DoC patients is still one of the biggest challenges of contemporary medicine.

One of the promising approaches to diagnose them was based on assessment of semantic integration processing. Sensitive to the semantic meaning, N400 is an ERP component observable in EEG signal over centroparietal brain regions. It is evoked by a semantic incongruence that occurs either in a sentence or between pairs of words.

The increase of the N400 amplitude in response to incongruent linguistic stimuli was observed in MCS, which suggests preserved language processing in these patients. On the other hand, some studies showed a lack of N400 in DoC patients and emphasised the difficulty of distinguishing MCS from UWS based on this component. One possible explanation indicates the presence of this component is dependent on the patient's intact structures, especially language networks. Thus, it can provide clinical information only on residual linguistic functions, and it is controversial if it can be an accurate indicator of preserved consciousness. It was confirmed by the results obtained in sleep studies and the ones with patients under general anaesthesia, where N400 was observed in unconscious patients.

Nevertheless, some research suggests that N400 can be seen as a good predictor of recovery - it correlates with the subsequent improvement of patients. In this study, we present a summary of research on N400 in the context of preserved consciousness.

# CLINICAL SESSION

**Hanna Nikanava**

## *Pharmacological treatment of migraine: mechanisms of action, efficacy and safety*

Migraine is one of the most common and debilitating neurobiological disorders. Unfortunately, this disease remains underdiagnosed and undertreated. Though pathophysiology of migraine is complex and not yet fully understood, numerous studies have provided evidence that the activation of the trigeminovascular system is involved into the mechanism of pain transmission (Negro & Martelletti, 2021). It was also determined that calcitonin gene-related peptide (CGRP), a neuropeptide expressed in trigeminal afferent neurons, plays a key role in the neurophysiology of migraine (Ho et al., 2010).

Nowadays, the 5-HT<sub>1B/1D</sub> receptor agonists, namely triptans, are considered as the standard of care for migraine acute treatment, exerting their antimigraine effects by blocking the release of CGRP from trigeminal afferent fibers (Clemow et al., 2020). However, triptan therapy doesn't help many patients to relieve from pain, disability, and migraine-associated symptoms (Cameron et al., 2015). Moreover, because of their vasoconstrictive properties, triptans are contraindicated in patients with uncontrolled hypertension and other cardiovascular and cerebrovascular diseases (Clemow et al., 2020).

New 5-HT<sub>1F</sub> receptor agonists, namely ditans, and calcitonin gene-related peptide receptor antagonists, namely gepants, as well as monoclonal antibodies binding to CGRP or the CGRP receptor have been developed as migraine-specific treatments devoid of vasoconstrictive properties (De Vries et al., 2020). In my poster I would like to describe the mechanisms of action of triptans and novel migraine treatment options and review the efficacy and safety of new pharmacological therapies.

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# CLINICAL SESSION

**Adamus S1**, Draps M2, Wierzba M3, Gola M2,4

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4Swartz Center for Computational Neuroscience, UCSD, San Diego, USA

## ***Resting-state functional connectivity in heterosexual males with Compulsive Sexual Behavior Disorder***

Compulsive Sexual Behavior Disorder (CSBD) is a disorder that was included in the 11-th edition of the International Classification of Diseases. It is characterised by inability to control urges to engage in various types of sexual behavior, which causes significant distress and interferes with patients' daily life. Brain mechanisms underlying this disorder are still poorly understood.

The aim of this study was to compare resting-state functional connectivity between patients with CSBD and healthy individuals.

The fMRI data from 52 patients with CSBD (mean age: 34.67, sd: 8.373) and 29 healthy controls (HC) (mean age: 34.93, sd: 8.831) were acquired in an MRI scanner during a 12-minute resting-state session (TR = 2000 ms, TE = 28 ms, slice thickness = 3 mm, FOV = 216 mm, GRAPPA mode). All subjects were heterosexual males. Data preprocessing and denoising were performed using CONN functional connectivity toolbox. Resting-state functional connectivity differences between CSBD and HC group were assessed using ROI-to-ROI analysis.

Our study revealed increased resting-state functional connectivity between left frontal orbital cortex (FOrb) and left insular cortex (IC) ( $t_{\max} = 2.13$ ,  $p_{\text{unc}} = 0.0366$ ,  $p_{\text{FDR}} = 0.5685$ ) in CSBD group compared to HC. CSBD patients also had increased resting-state functional connectivity between left inferior frontal gyrus (IFG) and bilateral supplementary motor cortex (SMA) ( $t_{\max} = 2.20$ ,  $p_{\text{unc}} = 0.0305$ ,  $p_{\text{FDR}} = 0.8366$  for the left SMA;  $t_{\max} = 2.70$ ,  $p_{\text{unc}} = 0.0086$ ,  $p_{\text{FDR}} = 0.1706$  for the right SMA).

To conclude, our results show altered resting-state functional connectivity in individuals with CSBD. Difference in functional connectivity between IFG and SMA was also observed by Seok & Sohn [1]. It was however task-related and decreased between right IFG and right SMA among CSBD group.

[1] Seok JW, Sohn JH (2020) Response inhibition during processing of sexual stimuli in males with problematic hypersexual behavior. *J Behav Addict* 9(1): 1-12. Funding: Polish National Science Centre OPUS grant 2014/15/B/HS6/03792, PRELUDIUM grant 2016/23/N/HS6/02906.

# CLINICAL SESSION

**Karolina Warzecha**

## *The conductor's score - the genes of the biological clock*

As early as the 18th century, researchers began working on the biological clock, checking how the mimosa would behave when placed in permanent darkness. Would its circadian cycles (folding and unfolding of leaves) still be the same despite the absence of the main time giver (zeitgeber)? The results were surprising, the plant still had a definite diurnal rhythm despite the lack of light. Similar research was done by Jurgen Aschoff in the 1950s. He confined a young doctor to constant light conditions for 40 days. It turned out that the subject's body also maintained its approximately 24-hour cycle. So how, if not through light, does our body continue to maintain a constant rhythm of internal processes? Every organism has an endogenous biological clock, which can be compared to a conductor who uses a baton to indicate the right time, intensity and tempo to the musicians playing in a symphony orchestra. Our organs are our musicians, and together they form a harmonious organism that acts in a specific, predetermined manner, influencing our mood and behaviour. We think we have power over it, but this is a false impression, because in an orchestra there is a place on a pedestal for only one person. And just as the conductor rules his orchestra, the biological clock rules us.

In that case, if it is not light and our will that controls the clock, then what is the cause of such a perfectly and regularly functioning mechanism? The answer is genes, the biological clock genes. They are found in every cell of the organs that function cyclically in our body and the place of supreme clock control - in the suprachiasmatic nucleus of the hypothalamus of the brain. The whole mechanism was discovered in the fruit fly, and I will present the complex mechanism of action on this model. It is based on the principle of 2 negative feedback loops and 1 positive feedback loop, which interlock with each other. Not following the guidelines of our inner guardian is associated with many health ailments ranging from very mundane, short-term to serious disorders of our body.

# CLINICAL SESSION

**Sandra Frycz**

## *Written in brain waves: using EEG to diagnose disorders of consciousness*

Coma, unresponsive wakefulness syndrome (UWS) and minimally conscious state (MCS) are disorders of consciousness (DoC) caused in most cases by brain damage like traumatic brain injury (TBI) or anoxia. Differentiation of such states is challenging, resulting in a high rate of misdiagnoses. In many cases, patients are not able to communicate or even move, yet remain conscious. Moreover, their ability to maintain arousal and show purposeful behaviour fluctuates in time. Thus, using only behavioural scales such as Coma Recovery Scale – Revised (CRS-R) to diagnose may not be sufficient. For that reason, current studies focus on neuroimaging techniques such as EEG, fMRI or PET. Bearing in mind that the assessments must be repeated multiple times for each patient, the use of fMRI or PET can be extremely expensive. In addition, it can be highly stressful for the patient. For these reasons, finding a method that's less expensive but as effective should be considered, which is why EEG protocols are widely applied by clinicians. EEG is a cheap, non-invasive method that is able to measure electrical activity of neurons, including neuron population oscillation or activity of neural networks which can correlate with the level of consciousness. Passive paradigms like resting-state EEG (rsEEG) or evoked auditory steady-state response (ASSR) can provide information not only about diagnosis but also about prognosis. rsEEG studies are used to evaluate neural activity in a patient's brain, without active involvement of patient in an experimental task. Characteristic patterns and frequency of brain waves during wakefulness can differ in people with DoC, giving information about their brain condition. Novel ASSR method is based on using chirp-modulation of periodic auditory stimuli which are presented to the patient. Auditory steady-state responses evoked by constant 40Hz stimulation may be used as a marker of thalamocortical network integrity, which is commonly disrupted in patients with DoC. Overall, using EEG protocols may play a huge role in reducing the number of misdiagnoses, contributing to better clinical care and higher chances for a recovery.

# CLINICAL SESSION

**Paulina Zegarska**

## *Amyotrophic lateral sclerosis - dysfunctions and therapeutic orientation*

Amyotrophic lateral sclerosis (ALS) is a very serious condition involving damage to the central and / or peripheral nervous system. The main symptoms include limb muscle wasting, muscle fasciculations, and a gradual loss of mobility. Treatment is only aimed at alleviating the symptoms and rehabilitating the patient.

A short 3-minute speech along with a poster presentation aims to introduce the participants to the problem. The poster is divided into several sections so as to be able to clearly illustrate the problem of providing as much information as possible in a short period of time. The presentation is a review work prepared thanks to the PubMed search engine and other sources found outside this search engine. In the first part, the overall picture of the disease will be touched upon. Then, aspects such as symptoms, diagnosis and treatment will be discussed. The next part of the poster presentation will focus on more specific questions.

The next section will be, in a way, a compilation of case reports related to ALS from 2020-21 (with PubMed after applying the “free full text” filter). The summary will be the buckle of the poster presentation. The poster is enriched with diagrams, illustrations, and various types of graphics in order to present the topic in the most accurate way.



# COGNITIVE SESSION

**Marta Chrustowicz, Łukasz Okruszek**

Institute of Psychology, PAS, Warsaw, Poland

## ***The impact of the transcranial current stimulation (tCS) over the right posterior superior temporal sulcus on neurophysiological markers of biological motion processing in patients with schizophrenia***

Social cognition (SC) is the ability to detect and process social information which is mediated by the “social brain” networks. The posterior superior temporal sulcus (pSTS), is one of the main nodes of the social brain which is activated e.g. during perception of biological motion (BM). Furthermore, decreased activity of this region during social information processing has been observed in patients with schizophrenia (SCZ) (Okruszek et al., 2017). It has also been suggested the dysfunction of this structure may be linked with decreased abilities to recognise social cues from the environment. Transcranial direct current stimulation (tDCS) which may cause an increase (anodal/atDCS) or decrease (cathodal/ctDCS) cortex excitability is proposed as a method that may improve social cognitive processes in SCZ. Thus, the aim of the current study is to examine the impact of the atDCS over the right pSTS on attention processes linked to the recognition of BM in SCZ.

26 SCZ were asked to complete biological motion perception task over two sessions preceded by either anodal or sham (control) stimulation while EEG signal was recorded. EEG event related potentials (ERPs) were reduced using principal component analysis (PCA) and a component corresponding to P300 potential was selected to measure the effects of stimulation. The results of the study indicated a significant interactive effect: a significant difference was observed between BM and SCR was observed after sham, but the same effect was not observed after atDCS. In sham condition, higher P300 amplitude in SCR than BM was observed. However, after atDCS this difference was no longer significant. The outcomes suggest that atDCS may have an impact on BM processing in SCZ. That implies that tDCS may be a potential for improving the social cognitive abilities in SCZ.

# COGNITIVE SESSION

**Aleksander Zębrowski, Laura Łępa, Wiktoria Jakubowska, Kinga Ciupińska, Marcin Koculak, Michał Wierzchoń, Michał Bola, Laboratory of Brain Imaging, Nencki Institute of Experimental Biology, Polish Academy of Sciences**

## ***Pre-stimulus alpha-band power predicts both subjective perceptual awareness and objective task accuracy***

At every moment multiple stimuli reach our senses, but only some of them gain access to consciousness. What mechanism determines whether a stimulus will be consciously perceived? Previous research has shown that spontaneous brain activity in the visual cortex, in particular the power of pre-stimulus alpha oscillation (7-14 Hz) affects whether the visual stimulus will gain access to consciousness or not. Specifically, when alpha power is high, detection of threshold stimuli is less likely. It is known that objective detection and subjective awareness of a stimulus might be in principle dissociated. However, in the study of consciousness, it is important to examine how alpha power determines not only objective detection but also the intensity of subjective experience.

In the conducted experiment a Gabor patch was presented in one of four peripheral locations, and in one of the four possible orientations (vertical, horizontal, tilted-left or tilted-right). A staircase procedure was used to adjust the contrast of a Gabor individually for each participant (N = 100), to obtain a 70% detection accuracy level. In every trial participants performed a detection task (i.e. reported in which visual quadrant the stimulus was presented), an identification task (i.e. indicated the stimulus' orientation), and rated intensity of their subjective perception using the Perceptual Awareness Scale (PAS). In the analysis the impact of pre-stimulus alpha power on tasks performance and subjective experience was modelled using linear mixed models.

The results showed that pre-stimulus alpha power occurring in the time window from -250 ms to -100 ms before the stimulus onset predicts both objective and subjective performance. We found that both accuracy and visual awareness decreased with an increase in pre-stimulus alpha power. Additionally, we found that the accuracy and alpha power effects on the PAS are independent.

Presented results bring important evidence into the ongoing debate showing that the power of low-frequency oscillations are indeed involved in a process of conscious perception of visual stimuli, in terms of both, subjective intensity of conscious experience and ability to detect and identify a stimulus. The results show that the pre-stimulus alpha power on a trial-by-trial basis is an indicator of the excitability of the visual cortex that determines objective task performance and subjective awareness.

# COGNITIVE SESSION

**Marcin Lewandowski, Monika Malon**

## *How subjective social status tricks our brains? An overview of neural, psychophysiological and behavioral evidence.*

The influence of subjective social status (SSS) is a relatively recent area of research in neuroscience. It has been reported that SSS affects multiple levels of one's life and well being, from physical and mental health to broadly understood cognition. This poster will provide an overview of the influence of SSS on both physiological and neurological markers as well as its further effects on socioeconomic decision-making. Individuals with lower SSS have been reported to suffer from higher levels of common stress markers such as heightened levels of cortisol and adrenaline, increased tonic electrodermal activity and lowered resting-state RMSSD, compared to individuals with higher SSS. They were also found to be aroused significantly longer by stressful events and needed more time for their stress markers to return to normal levels. Moreover, lower SSS has a particularly pronounced negative effect on mental health, including increased risk of depressive symptoms and impaired self-regulation abilities. Interestingly, during Ultimatum Game, low SSS subjects were more likely to accept unfair offers, yet at the same time they experienced more negative arousal markers measured by electroencephalography (EEG). Moreover, researchers have found ties linking SSS level with political beliefs, attitudes towards redistribution and towards individuals of opposed viewpoints. In conclusion the poster will provide an insight into potentially promising areas of further research in the field of SSS's influence on people's lives.

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# COGNITIVE SESSION

**Gregory Petrazzo**

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## ***Combination of Dasatinib and Quercetin improves cognitive abilities in aged Wistar Rats***

Introduction. Neurons and other glial cells have the potential to acquire senescent characteristics that could lead to defect in neuronal plasticity and alteration of cognition which negatively impact quality-of-life of elders. Eliminating senescent cells that accumulates with age, using senolytics drugs, has proven to be effective in alleviating symptoms of aged-related diseases.

Hypothesis : Combination of Dasatinib and Quercetin senolytics (D+Q) might prevent cognitive decline observed in aged rats. Objectives : Assess higher cognitive function of spatial working memory in young and aged Wistar rats treated with D+Q or vehicle. Methods : Young (3-month-old) and naturally aged male Wistar rats (18/22-month-old) were treated with either D+Q or its vehicle for eight weeks and tested in the active allothetic place avoidance task right after treatment and after a six weeks period of washing.

Results : We confirmed the cognitive decline of aged rats compare to young animals. We observed in aged but not in young rats treated with D+Q a reduction of entries, shocks and shocks to entrance ratio along with an increase of the maximum time avoiding the shock sector. Furthermore, D+Q treatment retains long lasting effects up to six weeks after treatment.

Conclusion : D+Q treatment alleviates some of the cognitive decline observed in naturally aged Wistar rats reflected by an improvement of spatial working memory. Our study brings new insights on the effect of D+Q senolytics in alleviating age-associated cognitive dysfunctions.

# COGNITIVE SESSION

**Emilia Kolada**<sup>1</sup>, Krzysztof Bielski<sup>1,2</sup>, Mateusz Wilk, Sylwia Adamus<sup>1</sup>, and Iwona Szatkowska<sup>1</sup>

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## ***Contribution of the centromedial and basolateral amygdala complexes to appetitive and aversive instrumental learning in humans.***

Extensive studies on animals have led to development of models of information processing in the amygdala during learning. The most influential model of fear conditioning postulates that associations between neutral cues and fearful events are formed in the lateral nucleus of the amygdala belonging to the basolateral amygdala complex (BLA) and defensive responses are mediated by the central nucleus of the amygdala belonging to the centromedial amygdala complex (CMA). However, relevance of this model to other types of learning has not been sufficiently explored. Here we used computational model of learning and MRI to investigate contribution of the BLA and CMA to outcome prediction and response of surprise during operant appetitive and non-fear aversive learning in humans.

Subjects (N=33) performed a reversal learning task, trying maximize the reward (in appetitive sessions) and minimize the punishment (in aversive sessions) by learning the probabilistic relationship between a neutral cue and a relevant outcome. Using behavioral responses and Rescorla-Wagner rule we calculated expected values and prediction errors, which were further applied to modulate the neural signal at the time of cue presentation and at the time of occurrence of an outcome, respectively.

During appetitive learning, significantly elevated BOLD signal to the expected values was found in the left and right CMA. The analysis of the BOLD signal generated to prediction errors showed activation of the bilateral CMA and the left BLA. Surprisingly, no activity in the amygdala was observed during non-fear aversive learning. Thus, the CMA appears to be involved in both processes, outcome prediction and production of a response of surprise. On the other hand, the left BLA is recruited specifically to surprise. These results shed new light on the information processing during appetitive learning in the amygdala in humans.

# COGNITIVE SESSION

**Ewa Wiwatowska**, Jarosław M. Michałowski

SWPS University of Social Sciences and Humanities, Poznań, Poland

## *Feedback processing in procrastination*

Procrastination is a tendency to delay tasks despite knowing that it may lead to negative consequences. Increased procrastination has been associated with higher fear of failure, maladaptive perfectionism and sensitivity to punishment, which suggests that high procrastinating students might be particularly susceptible to negative evaluation. In order to investigate this issue experimentally, we conducted a study in which high and low procrastinating students performed the reaction time task with positive or negative feedback, indicating whether participants managed to respond to the presented stimuli in the selected time window. During task completion EEG signal was recorded, in order to examine the event-related potentials linked to outcome anticipation (stimulus-preceding negativity; SPN) and allocation of attentional resources to presented feedback (P3). The obtained results showed that in comparison to low procrastinators, high procrastinating students presented larger SPN during outcome anticipation, but decreased P3 in response to feedback. These findings indicate that increased tendency to delay tasks is associated with higher anticipation of evaluation, but decreased allocation of attentional resources to process feedback information.



# COGNITIVE SESSION

**Gabriela Front, Małgorzata Paczyńska and Gabriela Puchała**

## *How brain can benefit from green tea?*

The neurodegenerative diseases number among the most serious and bothersome ones. The WHO statistics say that, considering current society aging processes, in 30 years there will be over 100 million people suffering from the Alzheimer Disease and about 12 million affected by Parkinson Disease. The origins of these diseases are still foggy and confusing. The drug therapies don't always work as they should and they are usually accompanied by many severe consequences such as liver damage. These facts may be really terrifying but there are ways of preventing our brains from symptoms of previously mentioned diseases. One of them, that has recently caught a lot of attention, is green tea. Known for almost five thousand years, green tea has been commonly used in China, Japan, India and Thailand. Many teated it as a traditional medication helping with most health conditions including bleeding, temperature control and digestion problems. Green tea became very popular and is prized not only for its taste but also plenty of properties beneficial for the human body. The present poster is a review of scientific research focused on the neuroprotecting properties showed by the molecules included in green tea, from the catechins displaying strongly antioxidant, anti-inflammatory and antipoptotic actions. Many issues related to neurodegeneration and green tea protecting affect on neurons has been brought up in our review. The main focus was placed on Alzheimer Disease and Parkinson Disease and their prevention that includes green tea consumption. Alfa-synuclein aggregation, neuroinflammation and neurons' apoptosis are the most common symptoms of the mentioned diseases. One of the most interesting chemicals is the EGCG catechin that can be found in green tea. Its neuroprotecting abilities are incredible. Apart from displaying antibacterial properties, it is also able to minimize the aggregation of alfa-synuclein which is responsible for the impairment of mitochondrial functions. There was also a mention of studies carried on mice that showed better motorcoordination and higher level of dopamine after using EGCG therapy.

# COGNITIVE SESSION

**Alba Casado**

Institute of Psychology , Jagiellonian University

## *Maintaining your native language when living abroad: an ERP study*

In the present study we explored whether migrants experience difficulties accessing their native language (L1) due to immersion in a foreign language (L2) environment. In particular, we explored if the L1 lexical access of migrants was disrupted in comparison with a control group of bilinguals living in the L1 environment. To this aim, we tested Polish-English bilinguals living in the UK (L2 environment) for at least 2 years and a matched group of Polish-English bilinguals living in Poland (L1 environment). Both groups of bilinguals named pictures in Polish (their L1), while we measured naming latencies and event-related potentials (ERPs). If the L1 lexical access is indeed compromised in migrants, we should observe longer naming latencies compared with the control bilingual group. Moreover, we should observe modulations of the naming P2 component —previously associated with lexical access— and/or the N300 component — previously associated with the ease of integration between the name of the picture and the picture recognition. The results showed similar naming latencies in both groups, indicating that at the behavioral level, migrants do not experience difficulties in L1 lexical access. In the ERP analyses, we found that the P2 component amplitudes were modulated by the language environment: higher amplitudes were evoked in the migrant compared with control bilinguals. Additionally, we found that the N300 was similar for both groups. In line with previous research, the higher P2 amplitudes could be interpreted as showing that migrants have more difficulty accessing L1 words. However, our component is unrelated to the naming latencies and shows a more frontal distribution than the naming P2 (centro-posterior).

Alternatively, the more frontal P2 component was previously related with the activation level of the previously used language (Branzi et al., 2014). Under this hypothesis, the higher amplitudes of the migrants compared with the control bilinguals may indicate that migrants used L2 before the task performance. However, this previous L2 use does not seem to affect the L1 lexical access, as shown by the lack of group differences in the response times and in the N300 component. In all, it seems that migrants adapted their language system to successfully access both L1 and L2, in such a way that behaviorally in picture naming latencies, no consequences are observed for L1 lexical access

# COGNITIVE SESSION

**Weronika Surdej**

University of Warsaw

## *The dark and bright side of the blue light.*

When it comes to life on Earth, one of the most important factors for living organisms is light. It's a main contributor in terms of regulation of circadian rhythmicity and controlling many biological mechanisms like vision, sleep-wake cycle, or brain activity not only in humans but also in other living creatures. Additionally, it has great influence on behavioural and psychological issues including cognitive processes. Numerous studies focus on blue light with wavelength in the range of 446-483 nm and its impact on circadian mechanisms with the aim of answering the question: is it destructive for homeostasis or can it actually improve our daily life performance? It's necessary to consider not only the length of the light but also duration of the exposure, intensity, and the time of the circadian day. It was proven that blue light influences sleep-wake cycle by regulation of melatonin secretion and has a connection to suprachiasmatic nuclei – a master oscillator that organises the peripheral clocks. Scientists addressed the fact that blue light has impact on alertness, cognition, mood, sleep, and phase shifting in circadian cycle. The point is – is it possible to determine if blue light is bad or good? Or is there some space for mutual agreement, a neutral ground? We live in the era of advanced technology. There are electronic devices with screens and diodes emitting short wavelength light all around us and it can lead to negative effects on the human body. On the other hand, exposure to blue light can be used in light therapies which are used in treatment of diurnal dysfunctions or depression. So let us take a closer look how blue light affects us.

# COGNITIVE SESSION

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## ***What the brain activity of cochlear implant users can tell us about our ability to localize the sound's source in space? The role of prestimulus oscillatory brain activity with respect to sound localization performance in cochlear implant users***

The ability to locate the source of a sound is critical to survival, i.e., it prevents being under threat when outside the field of vision. Further aspect in which spatial hearing is relevant, is human communication. Thanks to spatial hearing it is possible to distinguish among different sound sources in complex acoustic environments and selectively attend to sounds that interest us (Billig et al., 2019). Consequently, impairments and loss of hearing affect orientation in space, and communication.

In people suffering from sensorineural hearing loss, cochlear implants (CIs) can be used to provide a sense of hearing by artificially stimulating the auditory nerve. However, the efficiency of the coding of sound in CIs appears to be limited in multiple sound sources. The reason is the fact that in CI users, binaural cues are limited due to unsynchronized auditory input between both ears. Identifying the neural correlates underlying this deficit might help us understand the ability to localize sound in space in a healthy brain.

Rhythmic neural activity is ubiquitous across multiple spatial scales of the nervous system (Buzsáki, & Draguhn, 2004). Their associations have been revealed in distinct cognitive states and functions (Kahana, 2006). The role of oscillatory alpha-band (~8–13 Hz) activity has been linked with processes such as perception, attention, and memory, in a substantial body of literature (Bonfond & Jensen, 2012; Chaumon & Busch, 2014). Low alpha power reflects a state of high excitability and favors perception while high alpha power is related to an inhibitory state and gates perception. However, the first study which showed the presence of alpha-like rhythm that decreases in power following auditory stimulation pointed to a wider range of frequencies (Griffiths, 1997).

Thus, this study aimed to investigate the role of prestimulus oscillatory activity, encompassing a wider range of frequencies, including the alpha band. To examine the characteristics and topographic distribution of prestimulus oscillatory brain activity, in spatial sound localization, the oscillatory brain activity in cochlear implant patients had been recorded during the performance of a specially designed sound localization task. No significant differences were found in the prestimulus power spectrum in the frequency band of interest (2-30 Hz, in the step of 2 Hz) between trials, where spatial sound localization task was performed correctly and those in which was done incorrectly. Hence, this study failed to account for showing the role of the prestimulus oscillatory power in spatial sound localization. The objective of the present study was not only to investigate the accounts of oscillatory activity function but also to provide anatomical specificity that may unravel the neural correlates of spatial sound localization. To this aim source analysis (beamforming) was conducted, following the prestimulus oscillatory activity. Sources correlating with the ability to localize sound in space, active in the prestimulus period, were found in several unilateral cortical regions in the right hemisphere at 4 Hz, i.e., the precuneus, postcentral gyrus, cingulate cortex, posterior parietal cortex. It can thus be reasonably assumed that these cortical regions are crucial for spatial sound localization performance. These results demonstrate that prestimulus oscillatory activity should be accounted for in spatial sound localization. Brain areas involved in spatial sound

localization are not limited to a single region. What is more, the right hemisphere seems to play a vital role in spatial sound localization.

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# COGNITIVE SESSION

**Natalia Jakubowska**

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***Psychophysiological, but not behavioral, indicator of working memory capacity predicts video game proficiency.***

Visual working memory (VWM) is the ability to actively maintain visual information over short periods of time and is strongly related to global fluid intelligence and overall cognitive ability. In our study, we used two indices of visual working memory capacity: the behavioral estimate of capacity (K) and contralateral delay activity (CDA) in order to check whether training in a Real Time Strategy (RTS) video game StarCraft II can influence the VWM capacity measured by the change detection task. We also asked a question whether individual differences in behavioral and psychophysiological indices of VWM can predict the effectiveness of video game training. Sixty two participants (non-players) were recruited to the experiment. Participants were randomly assigned to either experimental (Variable environment), active control (Fixed environment) and passive control group. Experimental and active control groups differed in the type of training received. Training consisted of 30 hours of playing the StarCraft II game. Participants took part in two EEG sessions (pre- and post-training) during which they performed the VWM task. Our results showed that working memory capacity (K calculated according to the Pashler's formula) increases after training in both experimental groups, but not in a control group. We have also found correlation between average visual working memory capacity (calculated as K) and mean CDA amplitude no matter which group we are looking at. And, last but not least, we have found that we can predict the amount of improvement in the RTS video game by looking at the psychophysiological indices (CDA amplitude and theta band) recorded at baseline (before training), but only in the experimental group. We think that the strength of the psychophysiological indicator of VWM capacity might be a marker of the future success in video game acquisition.

# COGNITIVE SESSION

**Jakub Słupczewski**, Katarzyna Mańkowska (2), Jacek Matulewski (3) and Małgorzata Gut (1)

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## *The importance of variability in the performance of the number line estimation tasks in children with dyscalculia risk*

Dyscalculia is defined as a neurodevelopmental specific learning disorder leading to the impairment in mathematics. The aim of the study was to investigate number line estimation ability in children with diagnosis of dyscalculia risk. Fifty-two children (26 girls and 26 boys, mean age = 9.88) participated in the study. Thirty-two had diagnosis of dyscalculia risk (DR). They were compared to 20 typically developed children (TD). Participants performed two number-to-position tasks (they were asked to estimate the position of a given number on the empty number line, ranged 0-100 and 0-1000, respectively). Moreover, they performed two tasks (also with 0-100 and 0-1000 range), which required the verbal response determining an estimated number magnitude for the location that was indicated by an arrow on the line. The results of the verbal tasks showed the greater mean estimation error (EE) in DR group only in case of 0-1000 interval. In the number-to-position tasks the greater value of EEs in DR group both for 0-100 and 0-1000 interval were revealed. However, the most interesting result was found when focusing on values of variance within each group, not only in the context of variability descriptive statistics (SD and SEM) but also in the context of a variance showed in the scatter plots presenting individual data and the vast differences between EEs of particular participants. There was a considerable heterogeneity of EE values obtained in both groups and in each tasks, however in DR group this dispersion of individual results was clearly bigger. Thus, it could be concluded that Mean or even SD and SEM statistics do not manifest the real decrease of abilities in the case of some deficits, until we focus on individual data dispersion. It seems to be an important finding for example in regard to diagnosis of dyscalculia.

# COGNITIVE SESSION

**Marta Szymańska**

Nicolaus Copernicus University in Torun

## *The relationship between time and numbers processing in children, the effect of music education*

The processing of time and numbers has the shared neural basis and the cognitive mechanisms. Moreover, A Theory of Magnitude (ATOM) proposes that time, space, physical size, pitch and quantity are part of one general magnitude system.

The aim of the study was to investigate the relationship between the level of basic numerical abilities and the level of time perception as well as production (e.g. the reproduction of auditory stimuli duration) in children. Moreover, we calculated the correlation between the indices of both abilities in 2 group of participants: 1) children experienced in music education and 2) children from general population (control group).

A group of 65 children, aged from 7 to 10, participated in the study. Forty-five of them had no experience in music education (control group, C). They were compared to 20 children, who had access to music education (M). Each participant was examined with the use of two computerized tests to measure 1) the basic mathematical skills such as number comparison, numerosity assessment and number line estimation, and 2) time perception, production, and reproduction (also the rhythm processing).

The results of correlation analyses showed the several positive correlation between investigated abilities (e.g. between rhythm perception and number line estimation), that confirm the ATOM postulates, however only in the case of M group, while almost no expected correlation in the control group. Thus, it could be concluded, that music education may enhance the relationship between processing of numbers and time. It suggests that music education is beneficial for mathematical education in primary school.

# COGNITIVE SESSION

**Kinga Ciupińska**

Jagiellonian University

## *Differential Effect of Emotional Arousal and Valence on Subjective Duration Judgments*

Emotional effects on human time perception are usually attributed to arousal speeding up or slowing down the internal clock or to selective attention as an alternative mediator of these effects. This study explores how the emotional visual stimuli and its perceptual processing affect perceived duration of the time interval. Additionally, ERPs were measured to establish the neural basis for this effect. In this study 40 participants' performed a temporal bisection task while EEG signal was collected. Participants were presented with emotional visual stimuli of varying durations. They had to judge whether a stimulus was longer or shorter in comparison to baseline durations. Behavioral results revealed that emotional stimuli were rated as longer than neutral. Both valence and arousal dimensions of the presented stimulus influenced perceived duration, for example stimuli from high-arousal negative category were perceived as the longest. ERP analysis demonstrated that the amplitudes of P2, P3 and LPP potentials were more pronounced for high-arousal negative stimuli than other stimuli. For the N2 component, amplitude in this category was reduced but there were also significant differences between emotional categories. Taken together, these findings indicate that emotional experiences may increase temporal estimation and suggest that attention and arousal are both involved in timing processing, but on a different degree, depending on the emotional dimensions of the presented stimuli.

# COGNITIVE SESSION

Natalia Rutkowska, Michał Bola

Nencki Institute of Experimental Biology PAS

## *Forest before trees, or trees before forest? Detection of gist and objects during real-world scenes perception.*

What is the temporal hierarchy of visual perception? Some theories posit that perception unfolds in a local-to-global manner, with local objects being recognized first and then integrated to create the global meaning or “gist” of a scene. In contrast, the global-to-local accounts assume that the global properties of a scene are computed early and automatically, whereas objects are recognized at later stages. However, even though both sets of theories have clearly contradictory predictions, it is unclear which describes visual perception more faithfully, as few studies compared them directly.

Therefore, in the present study we directly compared the temporal aspect of the global and local level recognition (i.e. backgrounds and objects, respectively). We used images depicting either a natural (BN) or an artificial background (BA), with a single natural (ON) or artificial (OA) foreground object. Thus, scene images were either congruent (background and object from the same category) or incongruent (background and object from different categories). The experiment comprised four blocks and in each block one of the four scene characteristics was defined as a target. The goal of the participants (N=38) was to react by pressing a button if the briefly displayed (64 ms) image complied with this prespecified criterion. We used the three-way repeated measures ANOVA to investigate how the performed task (detection of background or object), the semantic congruency of an image (congruent or incongruent) and the target category (natural or artificial) affected detection accuracy (as indexed by  $d'$ ) and processing speed (mean reaction-times; RTs).

The results indicated that detection accuracy was higher for objects than for backgrounds and for congruent as relative to incongruent images. The analysis of interactions showed that participants responded more accurately to objects as relative to background and to natural as relative to artificial targets - but these effects were observed only in incongruent scenes. The analysis of mean RTs revealed that responses were faster to objects as relative to backgrounds, to congruent as relative to incongruent trials, and to natural as compared to artificial targets. Additionally, the analysis of interactions indicated that for background tasks and for artificial targets congruency of an image impacted the speed of its processing, so that incongruent images were processed longer than congruent ones. However, such an impact of congruency was not observed neither for object tasks nor for natural targets.

Our results stay in line with classical - local-to global - hierarchy of visual processing as they clearly show the temporal primacy of objects as relative to background in perception. Future studies are essential to investigate the specific factors underlying the visual hierarchy of real-world scenes' perception and its neurophysiological underpinnings.

# COGNITIVE SESSION

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## ***Investigating complex computational capabilities in the subcortical auditory system using Frequency-Following Response***

Previous studies have shown that the auditory cortex can extract complex contingencies between the incoming auditory stimuli. In this experiment we wanted to investigate whether the subcortical auditory system has similar computational capabilities.

A characteristic signal from the subcortical auditory system is the Frequency-Following Response (FFR). FFR is a component of the auditory evoked potential which mimics the eliciting stimulus signal. Using the so-called simple oddball paradigm, it has been shown that the grand-average power spectrum of the FFR to deviant stimuli was significantly larger than that observed in response to reversed-standard stimuli.

Here we used a much more complex paradigm where stimuli followed two different abstract rules based on the duration and frequency of the stimuli. A deviant stimulus was a particular sound that broke the rule established by preceding contingencies. There was also a control part in the experiment where the stimuli did not follow any of the rules. This paradigm has already been applied to show the complex computational capabilities of the auditory cortex.

We analyzed data from 30 participants from electrodes Fz, FCz, Cz and CPz. Data were re-referenced to the A1 electrode and filtered bandpass from 215 to 1500 Hz. After we preprocessed and epoched the data we computed amplitude spectra and made a grand average of the trials from the two durations. Mean spectrum value and signal to noise ratio in peaks in the frequencies of interest were analyzed by one-way ANOVA between deviant, reversed-standard and control trials.

Our results failed to show that in the case of the subcortical system there were any significant differences between the amplitude of the peak in the frequency of interest for the FFR elicited to the deviant, reverse-standard and control trials. This suggests that complex computations of auditory stimuli are not carried out in the auditory brainstem.

# COGNITIVE SESSION

**Alicja M. Olszewska**<sup>1</sup>, Dawid Drożdziel<sup>1</sup>, Maciej Gaca<sup>1</sup>, Wojciech Obrębski<sup>2,3</sup>, Jakub Kowalewski<sup>3</sup>, Agnieszka Widlarz<sup>3</sup>, Artur Marchewka<sup>1#</sup>, and Aleksandra M. Herman<sup>1#</sup>

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## ***When ears deceive you. Processing of auditory incongruence in musicians - preliminary results***

Errors are an inevitable part of the learning process. When playing music, musicians expect to hear a particular pitch as a result of a particular body movement, such as pressing a key on a piano. A sound of a different pitch is considered an error. These errors in auditory feedback are used by musicians to monitor the accuracy of their musical performance and facilitate learning.

We developed a highly ecological MRI-compatible keyboard instrument to investigate the processing of incongruent auditory feedback in pianists while playing in the MRI scanner. Eleven musicians (female, age 19-26) were scanned performing an altered auditory feedback task. In this task, musicians first listened to and then replayed various musical scales. In half of the trials, errors were simulated by replacing the auditory feedback of a single key with a sound corresponding to a neighbouring key (e.g. G -> G#). The musicians could not look at their hands, so they were not able to use visual feedback as a reference.

For neuroimaging data analysis, we created a full factorial (2x2) model using playing vs listening as one factor, and feedback condition (altered vs. correct) as the other factor. In the playing (altered auditory feedback) – playing (correct feedback) contrast, increased responses were found in multiple brain areas, including the left supramarginal/angular gyrus, left middle frontal gyrus, and bilaterally in the occipital and triangular parts of the inferior frontal gyrus (corresponding to the Broca's area in the dominant hemisphere).

These preliminary results suggest that the processing of unexpected auditory feedback (i.e. incongruent pitch sounds after a correct key press) involves the regions of the dorsal auditory stream in the left parietal lobe, which are related to musical performance and respond to musical training. The middle frontal gyrus has been associated with the shift of attention from inward to outwards, which might reflect the attention-grasping unexpectedness of the altered-feedback pitch. Finally, the activated regions in the frontal lobe are typically associated with the lexical processing of semantics. If confirmed, it would indicate that in musicians, the Broca's area role is broader, and involves the processing of structured auditory stimuli, not necessarily related to language.

# COGNITIVE SESSION

**Anna Żochowska**

Nencki Institute of Experimental Biology PAS, Warsaw, Poland

## ***Do surgical-like masks change our face perception? The processing of self-face, close other face and unknown face during the COVID-19 pandemic***

The COVID-19 pandemic has brought many changes to our everyday lives. Since March 2020, we have been instructed to take special measures of precaution in order to avoid virus transmission. Due to safety requirements, we have been confronted with faces (mouth and noses) covered by surgical-like masks on an everyday basis. This raises a question about how our brains process this kind of visual information, i.e. if it is processed like an unmasked face. In order to answer this question, in the current study we investigated the neural correlates associated with the processing of different faces (self, close-other's, unknown) with and without surgical-like masks. Our event-related potential (ERP) findings showed that covering faces with surgical-like masks had a similar impact on all of the faces. Amplitudes of P100, P300, and LPP were higher for this class of faces, thus at each stage of visual information processing images of faces with surgical-like masks benefited from the enhanced attentional processing. In addition, the prioritized processing of the self-face (with or without a mask) was observed, as revealed by increased P300 and LPP. N170 amplitudes were similar for both covered and uncovered faces, and despite covering the lower part of the face, a CLARA analysis localized the sources of brain activity in the fusiform gyri in both cases. The latter is in line with findings of some fMRI studies showing equally strong activations in this region both for entire human faces and parts thereof. Our results that show the activation sources in the fusiform gyri for covered faces may be viewed as evidence for the upper part of the face to be treated as a face already.

This research was funded by the National Science Centre Poland (Grant No. 2018/31/B/HS6/00461).



# COGNITIVE SESSION

**Krystian Dereziński**

Nicolaus Copernicus University in Toruń

## *Application of biometrics to optimize knowledge transfer*

In the field of education, the problem of adapting the content to the level of knowledge and skills of students continues to be a problem. It had its source in the economic dimension. Individual delivery of content requires enormous financial resources and in the case of educational institutions it was by definition impossible to achieve. Therefore, it is required to adjust the dynamics and difficulties of the knowledge transferred to the potential of the majority of recipients, which may result in difficulties in understanding the message in some users experiencing various types of educational problems or failure to meet the needs of acquiring knowledge by those with above-average abilities.

The topic presented here is the use of biometrics (with general consideration of passive brain-computer interfaces - pBCI) in adapting public utility facilities, and specifically cultural and educational institutions to the needs, possibilities, preferences and predispositions of the recipients of the presented content.

The system, which we are developing, allows to adjust the environment in real time to the current predispositions of the recipient, which on the one hand positively influences the transfer of knowledge, and on the other makes the presented content significantly more attractive to the user.

The modalities within which information about the user is collected are electroencephalography (EEG), visual object detection and eye tracking. The lighting, sound and content of the message (the content of the texts) are modified.

Our research is carried out in cooperation with museums in Poland (the Museum of the Origins of the Polish State in Gniezno and the Archaeological Museum in Biskupin).

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# COGNITIVE SESSION

**Aleksandra Lewandowska**

University of Bialystok

## *Bioelectrical brain activity & aggression*

Brain waves frequencies or their ranges correspond to certain mental states of a human being. The Delta waves are registered to the greatest extent in the sleep phase, during periods of reduced activity of pyramidal cells. Theta waves are associated with the extraction of information from memory and the ability to control the response to stimuli. Alpha waves dominate in states of rest and relaxation. Beta waves are related to the state of vigilance, external orientation, problem solving, logical thinking and focusing attention. The SMR rhythm is associated with intense prior thinking. For appropriate functioning, proper bioelectrical brain function is needed. In some cases, some waves dominate over others, which is an undesirable state. Research indicates a strong relationship between disproportions in the alpha and beta bands around the frontal as well as the high amplitude of P300 evoked potential in the fronto-temporal areas with the occurrence of aggression. Knowledge of these pathological states can help in the construction of effective supportive therapy for aggressive offenders.



# COGNITIVE SESSION

**Aleksandra Lewandowska**

University of Białystok

## *EEG Biofeedback in social rehabilitation*

EEG biofeedback is a method of learn by instrumental conditioning. It is assumed that rewarding the desired behavior increases the likelihood of this behavior in the future (Thompson, Thompson). It is based on monitoring the brain's electrical activity and consciously influencing its change. Neurofeedback therapy uses specialized equipment that reflects physiological processes that the patient is not aware of, and which are possible to consciously control. To measure these processes, an electroencephalograph is used, i.e. a device that gives the result of the EEG test. The EEG test measures the amplitude and frequency of the brain waves through small electrodes attached to the scalp with a conductive preparation. In this way, the device registers the bioelectrical activity of the brain reflecting the electropsychophysiological state of the patient. Although neurofeedback training is mainly used in the treatment of ADHD, Asperger's syndrome and attention disorders, some studies indicate its effectiveness in the treatment of aggression and hyperactivity (Denson, Miller, Pollock). Moreover, a study conducted on a group of adult criminal offenders, with particular emphasis on the tendency to aggression, violence, recidivism, the occurrence of dissocial personality, schizophrenia, attention deficit hyperactivity disorder (ADHD) and disorders related to the use of psychoactive substances showed a positive effect on cognitive abilities and improvement of attention (Fielenbach, Donkers, Spreen, Visser, Bogaerts). Similar observations were made on a group of juvenile offenders between the ages of 13 and 17 (Smith, Sams). The above-mentioned areas of effectiveness suggest that EEG biofeedback neurotherapy could be an important element for people undergoing the social rehabilitation process, which is the subject of the author's own research.

# COGNITIVE SESSION

**Krystian Dereziński**

Nicolaus Copernicus University in Toruń

## *The brain-computer interface in public space - an example of the application of the Steady-State Visual Evoked Potential (SSVEP) paradigm*

Steady-State Visual Evoked Potentials (SSVEP) is one of the most frequently chosen methods of communication with a computer within the Brain-Computer Interface technology.

SSVEP are visual steady state evoked potentials that appear in the electroencephalography (EEG) when the user focuses his attention on a visual stimulus that flashes at a constant rate.

In this presentation, I will describe the research that develops the SSVEP paradigm in a way that allows it to be used in the public space, so that it supports the interaction with the public space.

The presence of neurocognitive disorders significantly affects the deterioration of cognitive processes, such as memory, perception, attention processes and decision making. Especially today, due to the lack of treatment options for these disorders, like mild cognitive impairment (MCI), it poses a serious problem. In response to that we propose an adaptation of cultural centers and cultural spaces not only to healthy people, but also to those who may be completely excluded from it.

Overstimulation is the main problem faced by people with disabilities outside the home and, above all, during any attempt to function in the external environment. Eliminating the number of stimuli in contact with cultural institutions may turn out to be a real salvation for a wide audience.

The infrastructure solution proposed so far in the form of lifts or ramps for wheelchairs in institutions promoting culture, knowledge and science has focused only on meeting the needs of people with physical disabilities.

Unfortunately this is not a comprehensive approach, as it does not take into account the problems of many other excluded people. The solution we propose may contribute to greater availability of places to people who so far, due to the lack of system solutions, have been excluded from public spaces, such as museums and galleries.

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# COGNITIVE SESSION

**Olgierd Borowiecki**

Nicolaus Copernicus University

## *Cognitive skills are part of the declarative memory system*

This work is not meant to specify what cognitive skills are. The aim of this work is to point to the fact that the involvement of the hippocampus is a criterium for distinguishing cognitive skills from sensorimotor skills and that given this involvement, cognitive skills find themselves on an opposite side of this biologically meaningful boundary relative to the procedural memory. Given that, cognitive skills shall be included into the declarative memory, along with episodic and semantic memory.

Memory is being dissected in two since almost a century. Cognitive maps vs stimulus-response, declarative vs procedural; goal-oriented vs habitual; model-based vs model-free; flexible vs automatic; reflective vs reflexive; prospective vs retrospective; allocentric vs egocentric; roughly: “memory that” vs “memory how” (1-4).

The above divisions are pluralistically operationalised but all share one meaningful commonality - involvement or lack of involvement of the hippocampus (1).

There is one more division which should be recognised on this list - between cognitive and sensorimotor skills (5-8).

Although skills are being primarily categorised within procedural memory system, some skills have been called “cognitive skills”. The involvement of the hippocampus is a deciding factor whether a given experimental task would be categorised as an instantiation of a cognitive skill (1,6,8-10).

Thus, the “cognitive skills” shall be included in the declarative memory system, along with episodic memory and semantic memory (8).

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# COMPUTATIONAL SESSION

**Krystyna Mokrzycka**

*Is there a possibility of proving a person inculpable with neuroscientific evidence?  
Possibility of enhancing criminal procedure with neuroscientific tools.*

Proving a person guilty and – in consequence – punishing them accordingly is a rudimental aim of a trial. While there are some premises of culpability that do not raise many doubts, one of them has always been surrounded with a heated debate, and it is the premise of the perpetrator being unable to control their actions or to understand their meaning and consequences while committing a crime. The extent of this premise and its interpretation vary in any country that defines it and so do the ways of proving that the premise is actually present in the particular perpetrator's case. Lately a new way of proving the premise's presence has emerged – the neuroscientific way. fMRI scans and other techniques are being used to illustrate the person's overwhelming incapability of controlling or fully understanding their actions, despite suffering from no psychiatric disorders. Even a brain tumour can completely change a person's perception of reality. The point of this speech is to showcase the new, often unconventional ways of proving a person inculpable, while illustrating them with a few case studies from both U.S. and European Courts, and to eventually discuss the further future of using neuroscientific evidence in the courtroom and its possible impact on criminal procedure.



# COMPUTATIONAL SESSION

**Mikołaj Daraz**

Jagiellonian University

## *Artificial neural network in the classification of action potentials*

Machine learning is one of the most important method used by researchers and entrepreneurs in the problems of prediction, clustering and diagnosis of events. A special type of machine learning are multi-layer neural networks (MLP – Multilayer Perceptron), which are a very efficient and universally applicable tools. One of the interesting uses of MLP's is the classification of events (such as action potentials) in signal analysis.

The aim of this study was to create an artificial neural network capable of classifying action potentials: non-dopaminergic and dopaminergic in electrophysiological recordings. In vivo signals were recorded in ventral tegmental area and substantia nigra pact compacta by Intracellular Bridge Mode Amplifier. The key technologies used in this project are: Python, Tensorflow (machine learning library) and PyQt5 which were implemented for signal inspection and classification evaluation. Using simple data extraction techniques (with scipy Python's library), measurements of action potential shapes were obtained. With application of Tensorflow library, a neural network was created, capable of classifying action potentials based on the given measurements.

A high level of prediction of action potential labels was achieved, however, it should be noted that a small data set was used. This prompts further research to find better action potential features and improve network precision. It has been shown that using the widely known techniques of neural network architecture, the work of scientists analyzing the electrophysiological recordings can be remarkably simplified.



# COMPUTATIONAL SESSION

**Ada Kochlewska, Cezary Paziewski, Martyna Poziomska**

Physics Department, University of Warsaw, Warsaw, Poland

## *DEVICE FOR MEASURING SOFT ELECTRODES*

EEG measurements are crucial both in the world of neuroscience and clinical research. An indispensable element of the measuring device are electrodes, which convert ionic currents into electron current. This process will only be successful if the electrodes have the correct properties. It is assumed that the resistance of the electrode-skin system should not exceed  $5\text{k}\Omega$ . The resistance of the electrode should be as low as possible as the skin itself has a very high resistance. The resistance of the electrodes depends on many factors such as: the electrode material, their geometry, the electrolytes used on the electrode surface, frequency of the signal, and the current density at the electrode-skin interface. Nowadays, increasing attention is also paid to the fact that the electrodes should be soft and durable, as it allows to make long measurements without discomfort for the test subjects.

Here, to find the best soft wet electrodes with the lowest resistivity, we built the measurement device. The whole system is controlled by codes written in the Python language. Because of that, it is possible to change the numbers of parameters including the signal amplitude (voltage), sampling frequency, frequencies, time of the one measurement. Electrodes are placed in the piston which allows manipulating squeezing of the sponge.

Preliminary research shows that for a 1.5 cm height sponge electrode the lowest resistivity is when there is no squeeze and the auxiliary substance is saline (0.9%). Also, there are better results when the frequency is above 20 Hz. In future studies, we want to not only repeat the current measurements, but also test conductivity using other parameters.

# COMPUTATIONAL SESSION

**Kamil Lipiński, Piotr Bogorodzki**

The Institute of Radioelectronics and Multimedia Technology, Warsaw University of Technology

## ***Intra Voxel Incoherent Motion: Experiment Planning and Parameters Calculation***

**Introduction:** Intravoxel Incoherent Motion (IVIM) is a specific application of Diffusion-Weighted Imaging (DWI) in Magnetic Resonance Imaging (MRI). The IVIM concept has been proposed to estimate perfusion in tissues as blood flow is similar to a pseudo-diffusion process. Despite IVIM popularity, there is no easily accessible software allowing IVIM parameters calculation and analysis, nor is there a unique golden standard proposed for performing IVIM experiments, especially in low perfused tissues such as brain. In this work, a toolbox allowing IVIM-MRI parameters calculation (using three basic methods: using trust-region single step fitting, trust-region segmented fitting, and segmented grid search), visualization, and synthetic signal generation, is presented. The app was realized in MATLAB environment using AppDesigner for user-friendly GUI and Curve Fitting Tool functionalities for the fitting algorithm. An experiment, parameters calculation of synthetically generated IVIM data derived from different b-values and various SNR levels, was performed and results described.

**Results:** Designing and developing mentioned software allowed to perform an analysis of different sets of parameters for an IVIM experiment. Results indicate that selection of b-values influences parameter calculation quality. Application will constitute a basis for additional functionalities in further work.

**Conclusion:** It is crucial to plan b-values set for an experiment properly. It may help design a better study template, resulting in better parameter calculation quality and impact study results. Having software suitable for experiment testing may lead to improvement in study design. High SNR is required for good quality calculation for voxel based analysis.

# COMPUTATIONAL SESSION

**Sajad Shahbazi**

## *The role of benzo-dioxole-piperamide as an NF-Kappa B silencer and anti-neuroinflammatory agent*

**Background:** NF-kB contributes to the biosynthesis of various chemokines, cytokines, and enzymes. It plays many crucial roles in the upstream neuroinflammatory pathways. The phosphorylation of Ser32 and 36 residues of I $\kappa$ B subunit leads to disruption of the bind between NF-kB complex protein and the inhibitory subunit (I $\kappa$ B). The mentioned process activates the NF-kB complex to shift into the nucleus and binds to the chromosomal DNA. One of the most prominent targets to regulate the translocation of the NF-kB complex into the nucleus is the IKK- $\beta$  enzyme. Inhibitors may bind directly to the active site of the IKK- $\beta$  enzyme or suppress the gene expression of the IKK- $\beta$  enzyme and protect the integrity of the NF-kB complex and keep it in the inactive form inside cytosol.

**Methodology:** In the present study, we developed a novel NF-kB inhibitor encoded (D5) and investigated the efficacy of our druggable compound through various in silico, in vitro, and in situ tests. We have investigated the impact of D5 on the gene expression of the IKK- $\beta$  enzyme using rt-PCR. The enzymatic function of IKK- $\beta$  was indirectly monitored using a monoclonal anti-phospho-I $\kappa$ B- $\alpha$  (S32) to tract the radical phospho-I $\kappa$ B in the cytosol of microglial and astrocytic cells using western blot and immunocytochemistry techniques. The structural inhibition of IKK- $\beta$  by D5 and pharmacological properties of D5 were evaluated using in silico drug discovery tools such as Schrodinger suite 2011 and Accelrys discovery studio ver. 2.5. The statistical analysis was performed using Microsoft Exel 2007.

**Results:** The results indicated that D5 inhibited the IKK- $\beta$  enzyme in both genome and proteome. D5 demonstrated a significant reduction of the radical phospho-I $\kappa$ B- $\alpha$  in the cytosol of human microglia and astrocytes.

**Conclusion:** the brilliant protective effect on the NF-kB complex, the great pharmacological properties for oral administration, and lack of toxicity, made D5 the most prominent inhibitor of the NF-kB pathway for further studies on developing a potent anti-inflammatory and anti-neuroinflammatory agent.

# COMPUTATIONAL SESSION

**Anna Leśniewska 1,2, Urszula Górska 2,3**

1 Psychophysiology Laboratory & Consciousness Laboratory (C-lab)

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2 Institute of Zoology and Biomedical Research, Jagiellonian University

3 Department of Psychology, University of Wisconsin-Madison

## *Auditory EEG closed-loop system during slow-wave sleep*

The specific function of sleep is not fully understood today, but we know that it is essential to survive, and that quality sleep brings several benefits to our daily functioning - from cancer prevention (Huang et al. 2021) to cognitive performance (Kyle, Sexton, Faige, Luik, Jane et al. 2017). Based on signal registered from scalp electrodes, one 90-min cycle of human sleep can be divided into N1 light sleep, N2, and N3 deep sleep of non-rapid eye movements (NREM) and one phase of rapid-eye movement (REM) sleep. Characteristic feature of N3 NREM sleep is the presence of slow wave oscillation, as well as sleep spindles, which both are the markers of sleep stability (Bernardi et al., 2018).

Among many types of stimuli that were used to enhance slow wave activity, acoustic stimulation was suggested particularly efficacious (Tononi et al., 2010), especially when tones were played in longer blocks (Riedner et al., 2012). In this work, we aim to describe the idea for the new closed-loop system to be developed for our research, in which periodic auditory stimulation will be delivered to enhance slow-waves during NREM sleep.

The system will initialize its work in N2 NREM, detected with the predominance of K-complexes and sleep spindles as bursts of activity in sigma range (9-16 Hz). Next, the periodic auditory stimulation will be delivered for about 1000ms followed by the off-period, which will last until new slow wave will be detected. The main functionality of this closed-loop system will be real-time detection of slow waves by negative-going zero crossing in 200-800ms range. Computations will base on root mean square (RMS) of the frequency spectra.

When the RMS delta will exceed a predefined threshold (<4 Hz) and a wave of this frequency appears about 6 times in the 20 second window - the system flag such a situation as deep sleep. Detection of higher frequencies (alpha and beta), will be flagged as microarousal and hold the system. The closed-loop stimulation will enable to study brain response to periodic sounds evoking auditory steady-state responses during NREM sleep in a controlled manner. It will be also tested in a context of affecting architecture, and quality of sleep, towards future potential implications for people suffering from insomnia.

# NEUROBIOLOGICAL SESSION

**Weronika Bakun(1,2), Ksenia Meyza(1), Joanna Jedrzejewska-Szmek(1) and Ewa Kublik(1)**

1 Nencki Institute of Experimental Biology, PAS, Warsaw, Poland

2 University of Warsaw, Psychology Faculty, Warsaw, Poland

## ***A step toward understanding sensory disorders in autism - a pilot study of sensory sensitivity test in mouse model of ASD***

Autism spectrum disorders cover a wide range of abnormalities in both sensory processing and social behavior. While the relationship between these two areas is undeniable, the direction of their mutual interaction has been controversial (Hazen et al., 2014). To date, most research has focused on finding the causes of the social, communication, and cognitive difficulties found in autistics, having treated sensory disorders as secondary consequences of a period of limited social interaction. Currently, the hypothesis claiming that changes in sensory systems are primary to the developmental disorders and neurobiology of ASD is increasingly being considered (Robertson, Baron-Cohen, 2017).

Studies involving animal models of autism can have a significant impact on characterizing the nature of sensory disorders in ASD and, consequently, on the diagnosis and treatment of the disorder. They can point to particular changes in a brain structure, connectivity networks, or specific neurotransmitter pathways that lead to abnormal sensory perception. First, it is however essential to establish reliable behavioral tests verifying animals' sensitivity to sensory stimuli. We propose to use ecological, automated experimental cages (IntelliCage) with drinking corners labeled with sensory stimuli of varying intensity. We assume that animals with ASD-like traits would be less likely to select corners in which sensory stimuli are present.

In a pilot experiment we tested groups of mice from BTBR strain (a model of autism) and from a control (C57BL/6) strain. Mice lived in the IntelliCage and, in successive daily sessions, air puffs, rough paper and light flashes were applied in chosen corners. Preliminary results indicated that BTBR mice avoided corners with air-puffs significantly more than control animals and this difference increased with stimulus intensification. However, there were no clear differences in reaction to light flashes or rough paper. To better configure the proposed experimental model we plan to repeat tests on larger groups of animals and modify the location and timing of sensory stimuli presented in IntelliCage corners.

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# NEUROBIOLOGICAL SESSION

Oskar Wizła-Kubiak

## *Brain's orgasm - neurobiology behind it.*

From evolutionary point of view, sexual behavior is most important aspect of life, not only in human. Biologically goal of our existence is to maintain immortality of genes. Evolution developed precision and very effective strategy to sustain genetic material as sexual reproductive. Both sexes during sexual behavior, that has become more and more pleasurable over the ages, are aimed to conceive offspring. That pleasure was and still is a factor, that very often motivates individuals to find and encourage a partner in order to perform a sexual act.

Emotional and mental state that our species desire is pleasure. That experience is written as positive, satisfying, enjoyable, giving happiness and joy. In sexual behavior we strive to feel plenitude of pleasure – orgasm, which activate complex of brain structures and release neurotransmitters such as oxytocin. Orgasm and the pursuit of it is most enjoyable and satisfying faze of sexual act. Commonly is described as fulness of happiness, connection with a partner, mind-blowing or cleansing experience. During ejaculation is observed activation of ventral tegmentum area (VTA) which is obviously connected with reward system. We also see the activation of areas such as basal ganglia, claustrum, the anterior part of mesocortex and the anterior thalamic nuclei. However, activity in the hypothalamus was not observed. What is interesting in amygdala nuclei and entorhinal cortex there is disappearance of activity which suggest explanation for calmness and fearless emotional state of the body.

Of course, in moment of orgasm oxytocin is released. That hormone is connected specifically with abolishing fear, triggering caring and protective behavior as well as increasing trust to a partner. This peak of neurohormone have significant behavioral observation. Reflection of orgasm in the brain is not completely discovered. Connection between activations particular brain regions and neurhormones influence leave field to future research.

# NEUROBIOLOGICAL SESSION

Gabriela Vykysala

## *Behavior deconstructed? The use of DREADDs*

DREADDs (Designer Receptors Exclusively Activated by Designer Drugs) are engineered muscarinic G – protein coupled receptors that can be activated by the specific compounds in a lock-key way. These receptors are expressed in chosen neurons' populations as a result of viral induction or genetic transformation. Their crucial features are low affinity for endogenous ligands and very little constitutive activity. Two by far the most popular DREADDs are hM3Dq (excitatory) and hM4Di (inhibitory), while clozapine N-oxide (CNO) is a commonly used drug. This substance is a metabolite of antipsychotic drug clozapine and can be received with food/water, via injection or special minipump. The application of described technique enables the study of particular types of neurons and their role in different behaviors, especially associative learning, memory, reward – seeking behavior, feeding behavior, mood and pain. Thus, it offers the possibility of new treatments of behavioral disorders like schizophrenia or autism. Apart from its promising applications, the use of DREADDs has many other advantages like repeatability and good spatial resolution. It can also be used for longer studies (lasting for minutes or hours). What is more, the application is neither particularly expensive nor difficult. However, some disadvantages should be considered. Small, but significant fraction of CNO can transform into clozapine that binds to the endogenous receptors and has high affinity to DREADDs. At high concentration, not only might it cause some typical for this drug side – effects, but it can also cause effects not connected with CNO – DREADDs interaction itself. It poses the question about the reliability of the previous reports and encourages to improve current paradigms and find new designed receptors and complementary drugs with favored features (such as k-opioid-derived DREADD and salvinorin B). Moreover, the ligand can activate many different signaling pathways what makes drawing conclusion much more difficult. On the other hand, this truly represents the complexity of induced physiological changes. To conclude, despite this technique still being in the state of the early development, it is a great tool in behavioral neurobiology with promising therapeutical applications.

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# NEUROBIOLOGICAL SESSION

**Anna Goncerzewicz**

Laboratory of Emotions Neurobiology, Nencki Institute of Experimental Biology, PAS

## *Brain size, gut size and cognitive abilities: experimental evolution of energy trade-offs*

The enlarged brains of homeotherms bring behavioural advantages, but also incur high energy expenditures. The 'Expensive Brain' (EB) hypothesis posits that the energetic costs of the enlarged brain and the resulting increased cognitive abilities (CA) were met either by increased energy turnover or reduced allocation to other expensive organs, such as the gut. We tested the directionality of the evolutionary relationships between energy expenditures, brain, gut and CA using an experimental evolution model in which we subjected line types of laboratory mice to artificial selection on basal (BMR) or maximum (VO<sub>2</sub>max) aerobic metabolism - traits that are implicated in evolution of homeothermy, having been pre-requisites for the encephalisation and exceptional CA of mammals, including humans. High-BMR mice had bigger guts, but not brains. Yet, they performed better on the cognitively demanding tasks carried out in both reward and avoidance learning contexts. Furthermore, the high BMR mice had higher neuronal plasticity (indexed as the long-term potentiation, LTP) than their counterparts. Our data indicate that the evolutionary increase of CA in mammals was initially associated with increased BMR and brain plasticity. It was also fueled by an enlarged gut, which was not traded off for brain size.





# NEUROBIOLOGICAL SESSION

**Kinga Przybylska, Patryk Sambak, Anna Błasiak**

Department of Neurophysiology and Chronobiology, Institute of Zoology and Biomedical Research, Jagiellonian University, Cracow, Poland

## *Anatomical characteristics of the innervation of the midbrain interpeduncular nucleus by the nucleus incertus of the brainstem*

The preference for novelty is an evolutionarily preserved, fundamental survival mechanism that enables adaptation to a changing environment. Its dysregulation can lead to psychiatric disorders such as addiction, schizophrenia, and autistic behavior. Notably, the most common psychiatric diseases are related to stress and anxiety, and at the same time they predispose to the development of deficiencies related to the response to novelty. An important role in this plays a small structure of the midbrain - the interpeduncular nucleus (IPN). The IPN has dense bidirectional connections with stress sensitive brainstem structure nucleus incertus (NI). NI is the major source of the neuropeptide relaxin-3 (RLN3), a cognate RXFP3 receptors agonist, and RXFP3 receptors are found in IPN. RXFP3 activations have been shown to modulate social interaction in mice and impair the ability to express novelty preference in rats. It seems that the NI-IPN pathway is an important element of the novelty preference mechanism. In order to expand our knowledge on the neurochemistry of NI-IPN neuronal connections, especially in the context of RLN3 and RLN3/RXFP3 signalling, we used the tract-tracing method in which a retroAAV viral vector driving fluorescent protein mCherry expression, was injected into the IPN. Immunohistochemical staining was performed on sections of all RLN3 sources in the brain, and the retrograde-labelled relaxin-3-positive neurons were counted. The obtained results revealed that the most abundant relaxin innervation of the IPN comes from pars compacta of the NI, which suggests that RLN3/RXFP3 signalling within the NI-IPN pathway may underlie the influence of stress on the mechanism of novelty preference and exploration, however further studies are needed to verify this hypotheses.

# NEUROBIOLOGICAL SESSION

**Martyna Gorkowska**

Jagiellonian University

## *Anatomy, physiology and function of the nucleus incertus input to the ventral tegmental area of the rat*

Nucleus incertus (NI) is one of the mammalian brainstem structures involved in memory formation and generation of stress response and arousal. It performs its broad functions through GABAergic and glutamatergic innervation of many forebrain structures, including ventral tegmental area (VTA).

VTA is one of the primary sources of dopamine (DA) in the mammalian brain, which is a key neurotransmitter for motivation control and experienced based learning. However, we still do not know the details of the anatomy, physiology and function of the neuronal pathway from the nucleus incertus to the midbrain dopaminergic system. To fulfil this gap in our knowledge, we have performed:

- 1) extracellular single-unit recordings of DA neurons' activity of urethane anaesthetised rats, combined with optogenetic activation of NI neurons directly innervating VTA;
- 2) visualization of NI to VTA pathway by injecting retrograde and anterograde viral vectors carrying genes for fluorescent proteins;
- 3) real-time conditioned place preference/aversion (CPP/A) test, combined with optogenetic activation of NI to VTA neuronal pathway. Results of electrophysiological experiments revealed that there is a subpopulation of VTA DA neurons that is inhibited by activation of the nucleus incertus. Moreover, anatomical observations revealed that NI innervates monosynaptically DA neurons, localised predominantly in the ventromedial VTA. Furthermore, during the conditioning phase of the CPP/A test animals spent significantly less time in the chamber where NI was optogenetically activated. Interestingly, in the last day of the retrieval phase, when animals did not receive stimulation, they spent equal amount of time in both chambers of the apparatus.

Based on our experiments, we can conclude that NI is one of the nuclei exerting inhibitory control over the mammalian dopaminergic system. Moreover, behavioral and anatomical observations suggest that NI may be involved in signaling significance rather than the value of perceived environmental stimuli.

# NEUROBIOLOGICAL SESSION

**Zuzanna Mincikiewicz**

## *Characteristics of ultrasonic vocalisations of rat pups in a neurodevelopmental animal model of schizophrenia*

In isolation from the caretaker and the nest, rat pups emit separation vocalizations. Their function is to elicit caring behavior in the mother and thus increase the chance of survival. It has been shown that administration of the neurotoxin metylazoxymethanol (MAM) at 17 days of gestation causes behavioral, neurochemical and neuroanatomical disturbances in offsprings imitating schizophrenia symptoms. It is also known that prenatal exposure to MAM affects acoustic communication in rodents of all ages. However, there is little information about the influence of this factor on ultrasound vocalizations (USV) of pups and alterations occurring during the development of the animal. In order to assess the effect of exposure to MAM on USV of neonatal rats over a longer period of time, the maternal isolation test was conducted at the sixth, ninth and twelfth day of life. The results proved that rats exposed to MAM vocalize at a lesser level and produce longer calls compared to the control group. Moreover, it has been revealed that with the development of pups, the acoustic parameters of the USV modify, which results in a more complex and sophisticated signal repertoire. Over the course of time, pups emit shorter USV with a wider frequency range and increased modulation. These results confirm that MAM administration at an early stage of life causes deficits in acoustic communication, which may vary depending on the age of the rat pups. This study also offers greater insight into the evolution of vocalization in infant rodents.

# NEUROBIOLOGICAL SESSION

**Anna Bryksa<sup>1</sup>**, **Maciej Winiarki<sup>1</sup>**, **Joanna Borowska<sup>1</sup>**, **Joanna Jędrzejewska-Szmek<sup>2</sup>**, **Szymon Łęski<sup>2</sup>**, **Ewelina Knapska<sup>1</sup>** and **Alicja Puścian<sup>1</sup>**

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2. Laboratory of Neuroinformatics, Nencki Institute of Experimental Biology of Polish Academy of Sciences, Warsaw, Poland

## ***Diverse roles of the various neuronal classes in the prelimbic cortex in social bonding***

Although much is known about the neuroanatomy of social attachment, still its functional circuitry is poorly understood. Many studies indicate a crucial role of the prefrontal cortex (PFC) activity in sociability, however, the involvement of various neuronal classes in that process remains unexplored. We investigated the effects of the artificial manipulation of the activity of : pyramidal cells, parvalbumin-(PV+) and vasoactive intestinal peptide- expressing interneurons (VIP+) in the prelimbic part (PL) of the PFC, and evaluated its influence on the interest in spontaneous social interactions with conspecifics.

Animals were tested in Eco-HAB, an ecologically relevant, RFID-based system for assessment of sociability in group-housed mice, which enables continuous, individualized measurement of voluntary behavior. Using genetically modified mice selectively expressing Cre protein combined with the PSAM/PSEM-based chemogenetics approach, we performed a time-constrained, cell-specific manipulation of the PV+, VIP+ and pyramidal neurons activity in the PL and tested subjects social behavior during the 90 minutes following the systemic administration of the drug (PSEM) activating virally introduced artificial ligand-gated ion channels (PSAM). We show that PL-constrained inhibition evoked by the chemogenetics activation of the PV+ interneurons and inhibition of pyramidal neurons attenuates sociability by decreasing the time voluntarily spend together.

On the other hand, selective activation of the VIP+ neurons increases, however slightly, animals' propensity to interact with one another. Taken together, our data point to the diverse roles various neuronal classes play in the regulation of social bonding and thus lays the foundation for understanding the neural underpinnings of social bonding.

# NEUROBIOLOGICAL SESSION

**Karolina Protokowicz**<sup>1</sup>, Iwona Sirocka<sup>1</sup>, Ludwika Kondrakiewicz<sup>2</sup>, Marzena Stefaniuk<sup>1</sup>, Ewelina Knapska<sup>2</sup>, Leszek Kaczmarek<sup>1</sup>

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<sup>2</sup> BRAINCITY, Laboratory Of Emotions Neurobiology, Institute of Experimental Biology PAS, Poland

## *Early life immune activation changes psychosocial behavior of adult mice*

The development of the nervous system is strictly organized in time. Both, genes and the environment play pivotal roles in this process. A hallmark of neurodevelopment is exuberant synaptogenesis that when occurring improperly may produce neurodevelopmental disorders (NDDs), in which inflammatory reactions may act as a trigger. Nonetheless, NDDs are considered to have a multifactorial etiology, which is poorly understood. Such NDDs as Autism spectrum disorder, ADHD, schizophrenia or Tourette's syndrome are characterized by altered psychosocial behavior. Multiplicity and severity of symptoms result in different mental, physical and emotional consequences for the affected individual.

The aim of this study was to evaluate changes in psychosocial behavior of adult mice treated with lipopolysaccharide (LPS) in early life. LPS injections are commonly used to mimic bacterial infection in animal models. In terms of onset of exuberant synaptogenesis, postnatal day 7 (P7) in mice corresponds to 16th week of pregnancy in humans. On P7 mice pups were injected with either LPS or physiological saline. After 3 weeks, behavioral tests were conducted. To evaluate anxiety levels in various conditions, self-grooming test, marble burying test and elevated plus maze were used. Learning pace of animals has been tested in IntelliCage system. Sociability of mice has been assessed with Eco-HAB cage system. Additionally, the behavioral tests applied allowed for evaluating of animals' activity. Mice after LPS treatment were less anxious and more prone to taking risk. Additionally, LPS treated group was less active and spend more time together in the course of social test. Surprisingly, mice after LPS avoided social stimulus from unknown animals. During IntelliCage training the animals injected with LPS needed more time to learn new conditions. Therefore, early life immune activation induces alterations of behavior that resemble symptoms observed in human patients. Results from our preliminary studies are promising, but need further investigation into molecular mechanisms behind behavioral changes resulting from LPS injection.

# NEUROBIOLOGICAL SESSION

**Anna Radlicka**

Maj Institute of Pharmacology PAS

## *Effects of L-DOPA on cognitive symptoms in a mouse model of progressive parkinsonism*

Parkinson's disease (PD) is associated with a progressive degeneration of dopaminergic neurons of ventral midbrain. Symptoms of PD include motor impairment and cognitive dysfunction. L-DOPA, a dopamine precursor, is widely used in treatment and has mixed effects on cognitive symptoms. The aim of this study is to detect spatial gene expression changes within the forebrain areas that are linked to chronic L-DOPA treatment and its effects on cognitive functions measured in behavioral tests. Here, we present preliminary behavioral data obtained from the first group of tested animals. Adult TIF-IADATCreERT2 male mice were administered with tamoxifen to induce progressive loss of dopaminergic neurons. Starting from the 7th week after tamoxifen administration, their cognitive and motor functions were assessed in CatWalk gait analysis system, accelerating rotarod test and operant sensation seeking test. On the 14th and 15th weeks, when the parkinsonian phenotype was advanced, mice were given L-DOPA (18 mg/kg) or saline once daily and their cognitive abilities were tested in novel object recognition and OSS tests.

Here, we show how chronic L-DOPA treatment affects cognitive functions (short-term memory, sensation seeking) in a mouse model of progressive parkinsonism.

The presented results are a part of a study supported by the National Science Centre, Poland under research project "Spatial analysis of the effects of L-DOPA on gene expression in the prefrontal cortex in a mouse model of Parkinson's disease", no. UMO-2020/37/N/NZ4/03672.

# NEUROBIOLOGICAL SESSION

Sylwia Drabik,<sup>1</sup>

<sup>1</sup>BRAINCITY, Laboratory of Neurobiology, Institute of Experimental Biology PAS, Poland

## *Electrophysiological and morphological properties of rat interpeduncular nucleus*

Interpeduncular nucleus (IPN) is a small midbrain structure laying medially and ventrally to the ventral tegmental area (VTA). It is anatomically divided into seven subnuclei which also show some differences within their projections. IPN was described as a crucial structure in the processes of brain excitation, novelty recognition and opioids, nicotine and alcohol withdrawal. Despite its wide projections and variability of functions, little is known about its neurons apart from the fact that majority of them synthesise gamma-aminobutyric acid (GABA). Some authors suggest the presence of more than one electrophysiological type of neurons within the structure, but still, more research is needed to characterise them.

The focus of this work was to characterize a population of interpeduncular nucleus rostral subnucleus (IPR) neurons both on the electrophysiological and morphological levels. IPR is the unique subnucleus of IPN, positive for the tyrosine kinase A receptor (TrkA, nerve growth factor receptors), moreover, IPR together with IPC (central subnucleus) are the only two areas of IPN characterized with the presence of glutamatergic neurons.

Patch-clamp recordings combined with immunohistochemical staining and confocal microscopy were used to achieve the project's aim. Electrophysiological data obtained will allow to distinguish IPR neurons properties in comparison to other IPN subnuclei in the future. Moreover, Sholl analysis performed suggests the occurrence of two morphological types of neurons within IPR, however, the examined group was too small to perform statistical analysis and conclude this explicitly. Therefore, more research is essential to expand insight into the complexity of IPN.

# NEUROBIOLOGICAL SESSION

**Justyna Barut**

Maj Institute of Pharmacology PAS

***Exploitation of in vitro and in vivo new mice models induced by CRISPR/Cas9-lentiviruses intracerebral administration for study of presymptomatic Parkinson's disease.***

The study of neurodegenerative diseases, such as Parkinson's disease (PD), relies on animal models, which have certain caveats reflecting the complex aspects of the disease. They are mostly based on genetic basis of the disease or neurotoxines which do not reflect idiopathic forms. Using the CRISPR/Cas9 system, which has recently revolutionized the field of biotechnology, we have created a universal construct that will be used to generate transgenic animals in a more efficient. Single lentiviral, also, CRE-dependent vector with gRNA targeting the selected protein specific to the disease will be delivered by stereotaxic injection into to the brain of CRE-bearing mice in a specific cell populations.

We provide a standardized workflow for assessing mutagenesis in population of targeted neurons and astrocytes. The efficacy of this approach is demonstrated in vitro, in primary dopaminergic mice neurons and primary astrocytes cell cultures. We used gRNA targeted EGFP to visualize the effect of the mutation. Lentivirus transduction lowered EGFP expression by approx. 50% (shown by qPCR and fluorescent staining). We also confirmed it in vivo, making an attempt to create a pre-symptomatic PD model based on applying Cre-dependent lentiviral vector carrying the *Rrn3* deletion (transcriptional factor, TIF1-A) directly to locus coeruleus in DBHCre mice.

We obtained progressive degeneration of noradrenergic neurons restricted to locus coeruleus (up to 50-70% cell loss after 3 months from injection). The second model in which we study the effect of astrocytes in SN/VTA on dopaminergic neurons is the model in which we induce progressive degeneration of astrocytes through a vector carrying an *RRN3* deletion.

We found this approach to be more efficient than conventional gene knockout allowing targeting cells that would be difficult to differentiate and reducing time-consuming animal breeding in classic Cre/loxP mediated recombination.

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# NEUROBIOLOGICAL SESSION

**Magdalena Sobień**, Dobrochna Adamek-Urbańska

## *Histological comparison of Anabantoidei fish cerebellum*

The cerebellum, as part of the hindbrain, is present in all vertebrates except primitive chordates. It is responsible for motor-sensory functions and may differ in size, shape, and the cellular organization of its fragments. The most diverse and most numerous group of vertebrates is fish. As a result of the occupation of various environments, and thus adaptation to these habitats, the structure of the central nervous system (CNS) may differ. Another suborder with also distinctive behaviours is the Anabantoidei. In addition to their ability to breathe atmospheric air, they are characterised by unusual reproductive behaviour, which includes the construction of nests from air bubbles and mucous secretions by the males, the provision of food for the offspring by the parents, and the care or lack thereof of the young offspring compared to other fish species. The study aimed to provide a basis for further research into the basics of animal behaviour.

The histological comparison was made of the cytoarchitecture of the cerebellum in two representatives of the suborder Anabantoidei - the paradise fish (*Macropodus opercularis*) of the family Osphronemidae and the kissing gourami (*Helostoma temminckii*) of the family Helostomatidae. The specimens were whole brains fixed chemically, sectioned with rotary microtome into 6µm thick slices and stained with haematoxylin and eosin. The stained slides were analysed microscopically using a Nikon Eclipse NI-E microscope with a Nikon DS-Fi3 camera and NIS Elements AR software.

The comparative analysis of the cytoarchitecture of the cerebellum in both studied fish species showed a significant similarity in structure. Compared to mammals, the cerebellum of the Anabantoidei was not folded, and the final part of the dorsal part was slightly elongated. In all analyzed individual's histological layers of the cerebellum were found: white matter, granular layer and fine layer. Moreover, numerous large pear-shaped Purkinje cells constituted the ganglion layer between the molecular and granular layers.

The described structure of the cerebellum indicates high conservativeness of the structure of this part of the brain also within the Anabantoidei suborder.

# NEUROBIOLOGICAL SESSION

**Zuzanna Stawicka**

Jagiellonian University

## *It's never going only about food – the neurobiology of bulimia nervosa*

Bulimia nervosa usually begins in adolescence and is characterized by episodes of binge eating and purging which occur after eating a large amount of food. Individuals have also disrupting body image and sense of lack of control during eating. Bulimia nervosa over a lifetime can crossover to another eating disorder for example anorexia nervosa. Disproportions of morbidity due to gender are visible and women get ill more often than men. Research usually points out anorexia nervosa as a major eating problem, so understanding eating disorders is often general, despite evidence there are different reasons for these conditions. In my poster, I would like to summarize the foregoing knowledge regarding the neurobiology of bulimia nervosa and focus on comorbid diseases like mood disturbances, anxiety, addiction, and personality disorder, especially borderline personality disorder, which co-occur with bulimia.

Similarly like in patients with anorexia nervosa, patients with bulimia nervosa display atrophy of white and grey matter in some parts of the brain and disturbed running of reward and limbic system. Also in patients with bulimia, the insular cortex which is involved especially in interoceptive awareness shows a different response in relation to patients with anorexia and healthy controls. Patients with eating disorders also more often abuse alcohol and drugs, which can indicate a common cause of substance and food addiction. However, it is not known how food, without which we cannot live can become addictive. A lot of individuals with bulimia nervosa also suffer from anxiety disorders as well as other anxiety, and neurobiological causes of all of them, seem to overlap. It has been reported that bulimia and addiction, are marked by impulsivity, bad coping with fear and stress, and dysregulated functioning of the same neurotransmitters. In individuals with bulimia nervosa fear and stress can provoke binge eating, and remorse triggered by anxiety can yield compensatory behaviors like vomiting, excessive exercise, fasting, taking laxating drugs. As we can see, it is like a vicious cycle.

Treatment of bulimia is often unsatisfying because there's a high probability of relapse. Especially antidepressants e.g. fluoxetine and cognitive-behavioral psychotherapy are usually applied. Moreover, in today's world where people live under the pressure of body image and severe stress, it is supposed that in the future there will be higher morbidity of eating disorders. That's why the development of successful medication should be our crucial goal.

# NEUROBIOLOGICAL SESSION

**Kacper Witek\***, Karolina Wydra-Kolarska, Agata Suder, Kinga Gawlińska, Dawid Gawliński and Małgorzata Filip

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## *Maternal high-monosaccharide diet changes the adult offspring behavior*

In epidemiological studies, the excessive consumption of sugar added to food, containing monosaccharides (glucose and fructose), by mothers can affect the fetal programming of their offspring and have serious health consequences in young-adult life. In addition to metabolic changes seen as a development of diabetes and obesity, the maternal high-sugar diet may contribute to further behavioral changes such as mental diseases, including anxiety or emotional related disorders. The aim of our study was to examine the impact of maternal high-monosaccharide diets (HMD) on the adult offspring's behavioral responses. During the pregnancy and lactation periods, Wistar female rats consumed control (CON) or modified diets – rich in glucose (GLU) or fructose (FRU). After weaning, offspring of both sexes were separated and kept on the CON diet. Behavioral studies were started at postnatal day 59 and included the spontaneous locomotor activity (LMA), the novel object recognition memory (NOR), and the elevated zero maze anxiety-like behavior (EZM) tests. The results revealed that the maternal HMD intake changed the adult offspring's behavior and the effect was sex-dependent. Maternal GLU female (but not male) offspring showed an increase in distance traveled and ambulatory time compared to CON rats during the whole 2 h LMA recording. All-female offspring groups demonstrated the upward trend for a stereotypic time and in bursts of stereotypic movement, but not in resting time, which suggests abnormally repetitive behavior. In the NOR test, maternal HMD offspring did not exhibit a change in recognition index compared to CON after 1 h the familiarization phase, while 24 h after the familiarization phase male FRU rats showed an increase in time exploring the new object. Whereas, in the EZM test of maternal GLU offspring males revealed anxiety-like behavior characterized by a decreased time spent in open quadrants. Our results showed that prenatal exposure to the maternal HMD induces in adult rats both sexes behavioral changes. Different LMA behavior, anxiety phenotype, and memory changes in the maternal HMD offspring may predispose to development nervous or/and mental diseases, which requires further research. Also, the modified, maternal HMD emphasizes the key role of a properly balanced diet in development, especially in pregnancy and lactation. This work was supported by the Polish National Centre of Science grant number 2016/21/B/NZ4/00203 to MF and partly by the Institute of Pharmacology statutory funds. The authors declare no conflict of interest.

# NEUROBIOLOGICAL SESSION

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## *May addictive stimuli modulate adult neurogenesis in the Nucleus Accumbens?*

Neurogenesis is the formation of new neuronal cells. Possibility of this process taking place in the adult mammalian brain has been hotly debated throughout decades. Nowadays, the presence of adult neurogenesis is widely accepted in the hippocampus in humans and additionally in the subventricular zone in rodents (Owji & Shoja, 2020). Moreover, two recently published articles have identified a subpopulation of neuroblasts in the nucleus accumbens (NAc) (Chen et al., 2021; García-González et al., 2020). It has been suggested that these neural progenitor cells originate during an adult neurogenesis, which was mediated by external stimuli - chronic pain (García-González et al., 2020). NAc is a structure crucial for addiction-related behaviours such as drug craving and drug seeking (Scofield et al., 2016). It has been shown before that administration of cocaine can affect adult neurogenesis in the hippocampus (Noonan et al., 2008). Therefore, it is reasonable to suspect that addictive stimuli would modulate adult neurogenesis in the NAc.

In order to test that hypothesis a pilot study was conducted. The aim of the study was (1) to reproduce the identification of neuroblasts in the NAc and (2) to compare their quantity in mice that underwent addictive training with the mice from the control group. Herein, mice were exposed to either 7 days of cocaine or saline intraperitoneal injections. 2h after the last injection brains were extracted, fixed with paraformaldehyde and sliced on the vibratome. Finally, the protocol from the paper of Diego García-González et al. (2018) was adopted in order to confirm that neuroblasts or glia-like fibers may be present in the area of NAc. Immunohistochemistry staining was performed on the mice brain tissues. Slices with immunolabeled markers of neuroblasts (DCX) and radial glia-like fibers (GFAP) were imaged with a fluorescent microscope.

The results confirmed the presence of neuroblasts in NAc. Moreover, an increased number of neuroblasts was found in cocaine-treated animals. The results of this experiment are a message to continue further study to broaden knowledge about the origins of neuroblasts in the area of NAc.

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# NEUROBIOLOGICAL SESSION

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## ***Modulation of CX3CR1 as a promising strategy for neuromodulation after ischemia***

Stroke is a progressive neurodegenerative disease that results from the depletion of blood supply in a particular brain zone. As a result, nervous tissue, which normally requires big amounts of oxygen and glucose is damaged. Such pathological conditions might be mimicked ex vivo by utilization of oxygen-glucose deprivation (OGD) model on organotypic hippocampal cultures (OHC). One of the advantages of such an approach, while compared to standard experiments performed on isolated neurons, is the preservation of tissue complexity. It is extremely important because, apart from damage resulting from the inaccessibility of oxygen and glucose, the major cause of neurons death after ischemia is inflammation. In a present study, we aimed to show the role of CX3CL1 receptor – CX3CR1 in the immune response to stroke-mimicking OGD conditions. OHC undergoing OGD was characterized by significantly increased cell death (confirmed by both LDH assay and confocal imaging of propidium iodide binding in OHCs). Consequently, OGD conditions lead to disturbances in both actin and microtubular cytoskeleton organization. It was followed by changes in mechanical properties of tissue detected by the use of atomic force microscopy working in force spectroscopy mode. Since CX3CR1 is considered to be specifically expressed on microglia, in the next step, we eliminate microglia from OHCs by use of clodronate as well as modulate CX3CR1 receptor by use of AZD8797. For OHCs, under control conditions, both uses of clodronate and AZD8797 did not affect the viability of OHCs in a significant way. For OHCs undergoing OGD lack of significant changes in cell death level was observed when OHCs were pretreated by clodronate. Importantly use of AZD8797 decreased cell death of OHCs undergoing OGD. Such observation indicated the beneficial potential of modulation of CX3CR1 to decrease the negative impact on neuroinflammation after OGD. Consequently, the use of AZD8797 on OHCs pretreated with clodronate lead to reversion of this beneficial effect and observation of a similar level of cell death as in OGD OHCs and clodronate alone OHCs. This observation confirmed that modulation of CX3CR1 results in a beneficial effect. In the present work, we showed, that OGD performed on OHC is an effective model of stroke, which allows us to not only observe changes in OHC viability, but also more sophisticated changes like disaggregation of the cellular cytoskeleton and affected physical properties of cells. We showed that modulation of CX3CR1 decreased cell death levels in OHCs undergoing OGD. Importantly, we confirmed that the CX3CR1 receptor is specifically expressed on microglia because its modulation on OHCs pretreated with clodronate did not lead to the observation of a beneficial decrease in cell death after OGD.

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# NEUROBIOLOGICAL SESSION

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## *Responses of lateral habenula neurons to an aversive stimulus across alternating brain states of urethane anaesthetised rat*

The lateral habenula (LHb) is a glutamatergic epithalamic structure which, by signaling the negative value of events to the dopaminergic neurons, plays an important role in processes related to motivation and learning. Based on the responses of LHb neurons to aversive stimuli (AS) two populations within the structure can be distinguished. Additionally, differences in the response of LHb neurons to AS are correlated with the pattern and level of their basal activity as well as their spatial location. Neurons inhibited by AS display fast, regular firing and are clustered in the medial portion of LHb, whereas AS-excited neurons are slow, irregular firing and are uniformly distributed throughout the structure. LHb neurons relay information about AS to dopaminergic neurons of ventral tegmental area (VTA) by two pathways – either through direct excitation of GABAergic VTA neurons or via indirect pathway through GABAergic rostral-medial tegmental nucleus. Our previous study described a subpopulation of VTA dopaminergic neurons that respond to AS in brain state-dependent manner under urethane anaesthesia. Here, we wanted to verify if LHb neurons are also a subject of such changes which could possibly modulate VTA neurons' activity. To answer this question, we conducted in vivo, multichannel, extracellular recordings of LHb neurons' activity and their responses to the electrical footshock. All experiments we performed on urethane anaesthetised, male, adult rats. We observed, previously described in the literature, AS-excited and AS-inhibited LHb neurons, that responded to AS in the same manner, during both REM-like and non-REM-like brain state. However, we have also encountered a specific population of LHb neurons that changed their type of response to AS between brain states under urethane anaesthesia. Interestingly, we have not observed any unresponsive neurons which are described in many other studies as extensive population characteristic for LHb. Additionally, our results suggest that LHb neurons' basal pattern and level of activity differs between mentioned populations as well as between REM-like and non-REM-like brain state. This study extends the knowledge about subpopulations of LHb neurons' responses to the aversive stimuli and may shed some light on the relation between general state of the brain and neuronal processing of aversion. Funding: National Science Centre, PRELUDIUM 17 2019/33/N/NZ4/03011

# NEUROBIOLOGICAL SESSION

**Denisa Mihalj**

Biomedical Research Center of the Slovak Academy of Sciences

## ***The alterations in the hypothalamic gene expression of synaptic cell-adhesion molecules and inhibitory neurotransmitter markers in two autism-related mouse models***

Autism spectrum disorder is accompanied by altered social communication and behaviour with heterogeneous etiology. Modifications in several genes affecting hypothalamic neuronal cell populations have been suggested to underlie neurodevelopmental abnormalities associated with autistic symptoms. Alterations in neurogenesis and cell differentiation could be particularly important during early brain development characteristic with intense synaptogenesis. The aim of the present study was to compare gene expression levels of synaptic cell-adhesion molecules and inhibitory neurotransmitter markers in the hypothalamus in two autism-related mouse models at postnatal day 5. Shank3- and Magel2-deficient mice were used, both known for displaying autism-like behavioral symptoms. Moreover, Magel2-deficient mice are investigated for a deficiency of several hypothalamic neuropeptides, including oxytocin. We demonstrated significantly higher gene expression levels of cell-adhesion molecules Neuroligin 2 and Neurexins 1 $\beta$ -2 $\alpha$  in the hypothalamus isolated from Shank3-deficient mice compared to wild-type animals. Further analysis revealed lower expression levels of GABAergic markers (Gad65 and Gad67) in Shank3-deficient mice. In contrast, we found higher gene expression levels of Neuroligin 2 in Magel2-deficient mice, with no such changes observed in the gene expression of Neurexins 1-2, when compared to wild-type animals. Reduced expression of some inhibitory GABAergic markers (Gad65, vGat) and GABA receptor subunits (Gabra2), but not all (Gad67, Gabra1) was found in Magel2-deficient mice. Overall, it appears that autism-like conditions are associated with a disrupted balance of cell-adhesion molecules essential for the formation of inhibitory synapses, which is reinforced by observation of altered expression GABAergic markers. In a view of our previous results using these mouse models, change in the ratio of excitatory and inhibitory synapses in specific brain regions is suggested in neurodevelopmental conditions related to autism. Supported by VEGA 2/0148/21, VEGA 2/0155/20, and SK-FR-19-0015.



# NEUROBIOLOGICAL SESSION

**Emilia Gawron**

## *The Habenula as a target for depression treatment*

The habenula as a structure with the ability to modulate monoamine's level has recently gotten a great amount of attention from the scientists. Implementing these newest discoveries into depression treatment could help us improve its results. For example deep brain stimulation to habenula had been proven to help with depression symptoms significantly in both human patients and animal models. This structure is also possibly involved in antidepressant effect of ketamine, a drug which researches pin their hopes on for quick and effective treatment.



# NEUROBIOLOGICAL SESSION

**Marta Kołodziejak**

*You can read and understand this sentence thanks to these molecular mechanisms*

Working memory, amongst many things, allows us to store the words that we have just read in our head for just about the right time to be able to construct a whole meaningful sentence out of them. To be able to understand the title of this poster your brain has to remember for a short period of time all the words that the sentence contains and the order they are in. It is thought to be able to do so because of a phenomenon called short-term neural plasticity which may underlie the formation of working memory.

Neural plasticity is a phenomenon responsible for changes in the functioning and organization of neural circuits under the influence of previously experienced activity. Short-term plasticity lasts from tens of milliseconds to several minutes and is considered a fundamental process in information processing. Within short-term plasticity there are found facilitation, augmentation, and post-tetanic potentiation, which are responsible for increasing the signal transmission between neurons, and synaptic depression, which in turn is involved in weakening the synaptic transmission. The mechanisms of short-term plasticity include changes in the calcium channels and hence the calcium current, the intracellular concentration of calcium cations, which is related to, inter alia, local saturation of calcium buffers and changes in the properties and quantity of neurotransmitter vesicles. Moreover, the vesicles are suspected to exhibit some kind of heterogeneity, which may also have an influence on the activity-dependent plasticity. Some neural pathways are subject to the processes of strengthening and facilitating signal transmission, while others are dominated by silencing and depression. A single synapse is not limited to one type of amplifying or debilitating mechanism. Various processes can take place in it with different results at different times. Because of all these processes synapses are enhanced or depressed for an optimal period of time to keep nuggets of information intact prior to putting them altogether and giving them a meaning. All these complex phenomena are here eg. for you to read and understand this abstract.

# NEUROBIOLOGICAL SESSION

**Anna Janus**

## *Value- coding, salience- coding and alerting neurons*

Midbrain is the location of two major dopaminergic neurons sources - ventral tegmental area (VTA) and substantia nigra pars compacta (SNc). VTA neurons are heterogeneous in respect to its molecular and electrophysiological properties as well as their afferent and efferent connectivity. Diversity of dopaminergic (DA) neurons is also reflected in various functions assigned to them, ranging from control of movements through facilitating synaptic plasticity, up to engagement in motivational processes. The latter function is strictly connected with the ability of DA neurons to encode specific features of the received stimulus in their electrical activity pattern. The cutting edge paper by Wolfram Schultz showed phasic increases in activity of DA neurons in response to unexpected reward. These results were a cornerstone of Reward Prediction Error theory, which described DA neurons as the ones comparing expected and obtained outcome of the action taken. For a long time, a great number of evidence supported this theory, confirming uniform coding of value of the stimulus by DA neurons, with phasic increase in activity in response to rewarding and phasic pause in activity in response to aversive events. Nonetheless, body of studies have found that some DA neurons are excited by aversive events, which led to the conclusion, that the aspect of DA responses to motivational stimuli is more complex, than it was previously thought. Despite years of research these findings pointed out the need for expanding and refinement of existing theory about value coding. This debate led to division of DA neurons into two populations: one-coding value of the stimulus and second- signaling its importance (salience).

Even though this new theory explained some previously ambiguous results, the possible role of dopamine in motivational processing still remains unclear and debatable. A recent review of literature on this area proposed another type of dopamine signaling- alerting signals, which are hypothetically received by neurons encoding both motivational value and salience. Thus, such alerting signals may facilitate action of both types of DA neurons and thus supporting orienting, approach behaviors and cognitive functions, as well as promote preference to environments rich in sensory cues. Nonetheless, further experimental investigations are needed to establish how specifically alerting signals are reaching midbrain DA neurons. An important issue for future research is to fill the gaps in current knowledge about role of dopamine in motivation and goal-oriented behaviors.

# NEUROBIOLOGICAL SESSION

**Aleksandra Bartosz-Nowakowska**

## *Neurorehabilitation of the motor system through Functional Proprioceptive Stimulation*

For many years human neurological injury was thought to be static. Nowadays there are numbers of mechanisms that researchers have been working on to restore the nervous system. After an injury (spinal cord injury, traumatic brain injury, or stroke) the distal portion of an axon undergoes Wallerian degeneration while the proximal portion seals the damaged membrane to form an end bulb. Unfortunately, guidance cues are typically missing so once formed growth bulb fails to regrow. Damaged CNS neurons continue to degenerate – this process is known as secondary injury, which includes spontaneous and uncontrolled release of glutamate, damage to NMDA and AMPA receptors, loss of cell membrane potential. Neurogenesis occurs naturally in the healthy adult brain, however, the CNS does not fully self-repair after injury. To avoid further loss of neurons, that result in impaired movement neurorehabilitation should be undertaken. Several ways for therapy have been explored, including electrical stimulation and physical rehabilitation paradigms. The main goal of neurorehabilitation is to prime the motor cortex to enhance neuroplasticity and motor learning. One of the newly explored techniques is functional proprioceptive stimulation, which accurately mimics the natural sensorimotor activity of complex multi-joints movements. In the periphery, focal vibration entrains muscle spindle Ia-afferent firing rates at a one-to-one ratio. Sensory and motor systems are interdependent, therefore CNS reacts by initiating corresponding movements. FPS can be used from the very acute stage of trauma or injury from reducing spasticity, promoting motor activity and learning, facilitating muscle contraction for functional activity, obtaining an efficient motor control in functional activities, stimulating the proprioceptive system to using as a proprioceptive training to restore sensorimotor organization in the movement disorders. This method allows for faster recovery and restoration of the nervous system, thus bringing patients closer to normal motor function.

# NEUROBIOLOGICAL SESSION

**Olga Chaikovska**

Assistant Professor, Department of Human Physiology, National Pirogov Memorial Medical University, Ukraine

## ***Histochemical characteristics of nitroergic neuronal system during acute alcohol intoxication and nNOS blockage in the rat lateral septum***

Alcohol is a one of the most frequently consumed substances of abuse, which can cause addiction or alcohol use disorders (AUDs). Alcohol addiction leads to decrease of the life quality of patients and considerable economical burden. Neuronal mechanisms of addiction are intensively studied. One of the most important systems involved in this process is a brain reward system that includes lateral septum (LS). Additionally alcohol consumption changes activity of the neurotransmitter systems including the nitric oxide (NO). Recent studies for blockage of nitric oxide synthase (NOS) for cocaine addiction and late stages of AUDs demonstrated that a group of the substances known as blockers of NOS can be referred to as candidates for alcohol addiction therapy. The aim of our research was to investigate histochemical characteristics of NO-system in LS, its response to acute alcohol intoxication including or excluding neuronal NOS (nNOS) blockage with selective inhibitor – 7-nitroindazole (7-NI). This study involved three experimental groups of animals (control group (n=4), group with acute alcohol intoxication (n=4), group of nNOS blockage with acute alcohol intoxication (n=4)). For statistical analysis, one-way Kruskal-Wallis test was implemented to reveal differences between groups (Matlab, Mathworks). We have identified NOS-positive structures in LS consisting of big neurons, medium/small neurons and nerve fibers. Acute alcohol intoxication activated subpopulations of NOS-positive medium/small neurons and nerve fibers. Moreover, we determined that ethanol-induced changes can be blocked with selective nNOS inhibitor 7-NI.

# NEUROBIOLOGICAL SESSION

**Marta Zbysińska<sup>1</sup>**, Jakub Wojciechowski<sup>1</sup>, Hanna Cygan<sup>1</sup>, Jacek Rogala<sup>1</sup>, Tomasz Wolak<sup>1</sup>,

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## *Neuronal mechanisms underlying lip-reading*

Lip-reading is the ability that helps understanding speech in noisy conditions. It can also enable people with hearing impairment to improve auditory speech perception. This ability includes visual processing of phonemes (visemes), getting to their auditory representations and then analysing lexical meaning of the whole word. Analysis of lip-reading isolated, in the absence of auditory stimuli, gives us insight into neural correlates of this complex activity.

Brain regions involved in lip-reading that were consistently reported in studies are auditory association areas in superior temporal region, auditory–visual integration areas in the inferior parietal lobe, area of Broca in the left inferior frontal region and premotor areas in the precentral gyrus. There are also other regions described in the lip-reading network research, however these differ across studies. Brain activity during lip-reading can be influenced by factors like sex or level of lip-reading ability. It can also vary across different kinds of stimuli. Processing lexical stimuli involves other areas than non-lexical ones. There are also differences in lip-reading networks for short language elements like phonemes and longer ones, like words, phrases or even whole sentences.

First aim of this review is to summarise current knowledge on areas that are crucial for lip-reading ability and constant among individual differences, influencing factors and types of stimuli. Second aim is to identify factors that are responsible for differences between lip-reading networks. Supported by the Polish National Centre of Science grant number 2016/20/W/NZ4/00354.

## NEUROBIOLOGICAL SESSION

Katarzyna Bilińska, Christopher von Bartheld, Molly Hagen, Nicolas Meunier, **Rafał Butowt**

*Why do COVID patients lose their sense of smell, how many COVID patients lose their sense of smell, and can the SARS-CoV-2 virus travel from the nose to the brain?*

One of the most frequent symptoms of COVID-19 is the loss of smell and taste. Based on the lack of expression of the virus entry proteins in olfactory receptor neurons, we originally assumed that the new coronavirus (severe acute respiratory syndrome coronavirus 2, SARS-CoV-2) infect sustentacular cells and Bowman glands but does not infect olfactory neurons within olfactory epithelium. These data support the new hypothesis that viral infection of ACE2-positive cells present in the olfactory epithelium rapidly triggers an olfactory dysfunction in COVID-19 patients. However, some recent studies have reported viral infection in olfactory neurons, opening the possibility that the virus can directly infect the brain by traveling along the olfactory nerve. We here review the current evidence for an olfactory route to brain infection and conclude that the case for infection of olfactory neurons is weak, based on animal and human studies. Our data points to alternative routes to the brain, other than along the olfactory projections, such as the route along terminal nerve. COVID-19 patients can be assured that loss of smell does not necessarily mean that the SARS-CoV-2 virus has gained access to and has infected their brains. Moreover, the prevalence of chemosensory dysfunction in patients with COVID-19 varies greatly between populations. Our epidemiological analyzes indicate that populations infected predominantly with the G614 virus had a much higher prevalence of anosmia compared with the same ethnic populations infected mostly with the D614 virus strain. We conclude that the D614G mutation is a major contributing factor that increases the prevalence of anosmia in COVID-19, and that this enhanced effect on olfaction constitutes a previously unrecognized phenotype of the D614G mutation. The new virus strains that have additional mutations on the background of the D614G mutation can be expected to cause a similarly increased prevalence of chemosensory dysfunctions.