Detailed information on
"Methods and Techniques in Neuroscience"

Methods and Techniques in Neuroscience combine the lecture Methods and Techniques in Neuroscience (VO 902557) and the practical course (UE 902558) in one united block starting mid-March and ending mid-April. Each day the students will be involved from 9:00-12:00 and 13:00-16:00. Lab Coats are required.

Due to the scheduling conflicts of the involved researchers, the order of the Blocks may change.

Abbreviations used for the location of the course:
CBR Center for Brain Research, Spitalgasse 4, 1090 Vienna, large seminar room first floor
CIN Clinical Institute of Neurology, AKH, Level 4J (meting point Leitstelle 4J)
Neurology BT86, AKH and **Hochfeld NMR**
Block 7 Schwarzspanierstraße 17, I

Neuroanatomy (CIN)
Lecturer: Romana Höftberger, Ellen Gelpi Mantius

General goal and basic knowledge: Based on autopsy cases of the Neurobiobank of the Medical University of Vienna, the students gain insight in the gross anatomy of the human brain, which will foster a broader understanding of the structural and functional organisation. Moreover the students will see different diseases affecting the central nervous system, including tumors, inflammation, and degenerative as well as vascular lesions.

Practical skills:
•Hands-on recapitulation of the gross-macroscopic organization of the human brain.
•Detailed inspection and characterization of diseased and non-diseased human brains.
•Correlation of anatomic changes and/or lesions to functional deficits. Diseased states cover various developmental, vascular, neoplastic, inflammatory and neurodegenerative pathologies.

Block 1: Immunohistochemistry and in situ hybridization in neuroscience (CBR)
Lecturer: Igor Adameyko, Polina Kameneva,
Elena Maria Kastriti

General goal and basic knowledge:
The students will get an in-depth understanding of using histological techniques and in situ hybridization (RNAscope, BASEscope, Hybridization Chain Reaction) for profiling the cells types and gene expression patterns in CNS and PNS. In this part of the course the students will learn the basic techniques for preparing the tissue for immunohistochemistry and in situ hybridization: both paraffin-embedded and frozen tissue, followed by the specifics of chromogenic and fluorescent revealing techniques. The theoretical part will also include specifics of imaging of different types of immunohistochemical and in situ preparations with the relevant post-processing and analysis techniques.

Practical skills:
Students will learn a variety of histological techniques such as cutting paraffin-embedded and frozen tissues, preparation of the sections for immunohistochemical staining, the steps of immunohistochemical staining, and revealing protocols (chromogenic revealing combined with H&E and multiplexed fluorescent revealing). Students will observe the setup for RNA/BASE scope protocols. Students will practice the specific settings for imaging of various sample types with confocal microscopy and learn about relevant ways of analysis.

Block 2: Neurophysiology (CBR)
Lecturer: Ruth Drdla-Schutting, Roni Hogri

General goal and basic knowledge: The students should understand membrane properties and basic mechanisms underlying the communication between neurons: resting membrane potential, action potential and synaptic transmission.

Practical skills:
Realistic computer simulation of the electrical behaviour of a nerve cell; Nernst and Goldmann equation, respectively, are used to understand the flow of ionic currents through membranes and the resulting formation of cell potentials; Excitatory and inhibitory synaptic transmission, NMDA-receptors. Practical training: Patch-clamp recordings from spinal neurons in a slice preparation (in current- and voltage-clamp mode); Measurement of action potential firing patterns.

Block 3: Pathobiology (CBR)
Lecturer: Johannes Berger, Isabelle Weinhofer

General goal and basic knowledge:
To learn how innate immune cell properties can determine brain inflammation, neurodegeneration and remyelination.

Practical skills:
MACS-bead isolation of specific cell types and determining their purity using FACS; in vitro differentiation of monocytes to macrophages; generation of lipophages by myelin phagocytosis, analyzing the morphological changes using microscopy, determining the phagocytosis rate using FACS; isolation and reverse transcription of RNA into cDNA; detection and quantitative analysis of gene expression using quantitative RT-PCR.

Block 4: Receptor Structure, Function and Pharmacology (CBR)
Lecturer: Petra Scholze, Margot Ernst

General goal and basic knowledge: The students should understand the basic mechanisms of receptor – ligand interaction and the techniques, which can be used to study these interactions.

Practical skills:
In the first part membrane suspensions from mouse brains will be prepared, and a radio ligand will be bound to the benzodiazepine binding site of $GABA_A$-receptors. Data will be subjected to computer analysis.
In the second part, experimental structures of $GABA_A$ receptors will be analyzed with different computational tools and aspects of computational structure guided drug design will be discussed.

Block 5: Synaptogenesis and Optical Imaging (CBR)
Lecturer: Ruth Herbst, Inga Koneczny, Klaus Becker, Hans Dodt

This Block combines two different topics:
Synaptogenesis (Lecturer: Ruth Herbst,
General goal and basic knowledge:
Introducing the neuromuscular synapse as a model system. Focus on the molecular mechanisms that lead to synapse formation in particular accumulation of neurotransmitter receptors during postsynaptic differentiation.

Demonstrations and practical exercises:
- Acetylcholine receptor clustering assay (differentiation and induction of muscle cell cultures, acetylcholine receptor staining, fluorescence microscopy)
- Purification and analysis of acetylcholine receptors (acetylcholine receptor labelling and pull-down, generation of cell lysates, SDS-PAGE, Western blotting, quantification of acetylcholine receptors)
- Tutorial on image analysis using ImageJ
Light sheet microscopy of cleared specimens (Lecturer: Klaus Becker, Hans-Ulrich Dodt)

General goal and basic knowledge: Introduction to microscopy in general and especially to light sheet microscoy. Introduction to tissue clearing.

Demonstration and practical excesis: Demonstration of cleared specimens and of recording of fluorescently labelled specimens with light sheet microscopy and of image processing

Block 6: Molecular Neuroscience (CBR)
Lecturer: Roman Romanov,

General goals and basic knowledge: The students should obtain insights in protein structure, receptor-ligand interactions, structural similarity between homologous proteins, different transgenic mouse models and their experimental uses.

Practical skills:
PDB database searching, viewing PDB structure files with the VMD viewer, aligning protein sequences with the ClustalX software, familiarity with fixed brain tissues and its processing, work with reporter mice (GFP labelled neurons) and understand imaging (fluorescence and confocal laser scanning).

Block 7: Mouse models/Behavior analysis (Schwarzspanierstraße 17, I)
Lecturer: Daniela Pollak

General goal and basic knowledge: The students will learn the basics about the pathophysiology of psychiatric disorders and available mouse models and behavioral tests. They will be introduced into behavioral phenotyping methods, including considerations for the design and practical execution of these experiments as well as behavioral pharmacological approaches. Pitfalls and limitations will be analyzed and discussed. Background about the framework for application for animal licenses (“Tierversuchsantrag”) will be provided.

Practical skills: - Behavioral tests in mice, including neurological observational battery, Open Field, Rota Rod, Elevated Plus Maze, Forced Swim Text, Tail Suspension Test, Social Interaction Test and Fear conditioning and respective analysis
- Evaluation of mouse maternal behavior and pup ultrasonic vocalizations and analysis
- Data evaluation and interpretation.
- Theoretical design of a project involving mouse behavioral analysis and application for an animal license ("Tierversuchsantrag")

Block 8: Cognitive Neurobiology (CBR)
Lecturer: Hugo MalagonVina,, Paul Anderson, Balint Lasztóczi, Claudia Espinoza

General goals and basic knowledge: The students should become familiar with the planning of a complex behavioral/electrophysiological experiment and subsequent histological analysis, and obtain insight into network dynamics and behavior-dependent neuronal correlates.

Practical skills: Rodent brain slicing, immunohistochemistry in free-floating brain slices, evaluation of multiple fluorescent IHC experiments, preparation for electrophysiological single-cell and neuronal population recordings, using stereotaxic instruments for orientation, behavioral experiments in real- and virtual-environment, use of light microscope and neuronal reconstruction for identifying cell types.

Block 9: Magnetic Resonance and Brain Stimulation (CBR, Neurology and Hochfeld NMR)
Lecturer: Roland Beisteiner,

General goal and basic knowledge: The students should understand the principles of signal generation with magnetic resonance technologies. Further they should learn which are the major magnetic resonance effects which can be used for neuroscientific investigations and clinical research and what are the benefits of ultra-high field systems. This concerns particularly the monitoring of human brain function, the monitoring of structural brain changes (neuroplasticity) and the monitoring of brain pathology.

Practical skills:
The students will be introduced to practical handling of functional magnetic resonance imaging (fMRI) experiments and they will perform analysis of fMRI data with standard software packages like SPM or FSL.