CircProt
Synaptic circuit protection in AD and HD: BDNF/TrkB and Arc signaling as rescue factors

Background:
Regulation of synaptic plasticity by brain-derived neurotrophic factor (BDNF) is crucial for brain function, as it pilots adaptive changes in neuronal networks. Pathological changes in BDNF availability and tropomyosine related kinase B (TrkB) signaling are therefore among the most relevant pathomechanisms in neuro-degenerative disorders (NDs). Huntington´s disease (HD) and Alzheimer´s disease (AD) are both strongly associated with BDNF related impairments. While BDNF is recognized as an endogenous protective factor in both diseases, the development of therapeutic strategies has been hampered by the lack of knowledge on BDNF transport and release, and on BDNF/TrkB downstream signaling networks in these NDs.
We propose that BDNF/TrkB signaling via Arc function is key for the management and treatment of synaptic dysfunction and neuronal degeneration in AD and HD. This project aims at identifying novel combinatorial and synergistic strategies to alleviate AD and HD related impairments based on regulation of TrkB and its downstream signaling cascades. In addition, since BDNF is an important TrkB upstream regulator, mobilization of endogenous BDNF synthesis, transport and release are investigated for their therapeutic potential.

Hypothesis and Strategy:
The key protective mechanism addressed is enhancement of endogenous BDNF/TrkB signaling induced by: enriched environment (EE), voluntary running (VR), or application of drugs that enhance BDNF expression (fingolimod, fluoxetine), BDNF vesicle transport (tubastatin and cysteamine), or TrkB signaling efficacy (cholesterol, fluoxetine), respectively. Advanced molecular imaging, synapse electrophysiology, spine density/morphology, biochemistry, and behavioral testing combined with computer assisted realistic neural network modeling, are used to determine optimal therapeutic strategies. The parallel analysis of AD and HD associated synaptic circuit dysfunctions and its drug-induced rescue, including modulation of neuro-inflammatory responses, will help us to identify common and divergent cellular pathways. Disease models used: 6-7 months old APP/PS1 mice as AD model. Long-term microfluidic cultures (>80 days) of human HD patient iPSC-derived neuronal networks as HD model.

Results:
AD: Untreated 6-7 months old AD mice held in standard housing (SH) conditions showed reduced LTP, and reduced spine densities in CA1 pyramidal neurons of the hippocampus, as well as impaired context fear conditioning (CFC) and Morris water maze (MWM) performance compared to their wild type littermates. Biochemical analysis of hippocampi obtained from untreated AD mice suggests alterations in translational control and Arc expression in APP/PS1 mice. These changes appear to be specific to CYFIP1/FMRP- and eIF4E-pathways that are downstream of TrkB-ERK-MNK signaling. Start of treatment with fingolimod (FTY720; FDA approved drug against Multiple sclerosis) after the onset of disease symptoms reversed hippocampal LTP, spine, and memory deficits in the AD mice. This fingolimod induced rescue occured independent of increased BDNF protein expression and downstream TrkB signaling but was mediated by anti-neuroinflammatory actions on microglia and astrocytes.
Providing 4 months old AD mice with VR opportunities (running wheels) led to increased hippocampal BDNF levels and rescued the LTP, spine, and memory deficits. Similarly, but slightly less effective, also EE ameliorated synaptic deficits of AD mice. Computational modeling of BDNF/TrkB dependent spike timing-dependent synaptic plasticity (STDP) in CA1 pyramidal neurons revealed a comprehensive understanding of BDNF-dependent mechanisms of synaptic plasticity that are affected in the CA1 region of AD mice.
HD: Neuronal differentiation of 3 different lines of HD patient derived iPS cells (iPSCs) was established. Differentiation to either striatal or cortical neurons was obtained using distinct protocols and showed a decline in BDNF protein expression and TrkB signaling that was previously observed for HD affected brains *in vivo*. Microfluidic chambers allowing co-culture of striatal and cortical neurons in separate compartments that are connected by channels, in which cortico-striatal synapses can form, were used to quantify BDNF vesicle transport and TrkB signaling. When culturing HD mouse derived striatal and cortical neurons in this system, we observed reduced BDNF transport in cortical axons and reduced pERK activation. Biochemical analyses revealed that fluoxetine and cholesterol binding to TrkB receptors modulates efficacy of TrkB signaling, pointing to a promising potential role of these TrkB signaling modifiers to counteract impaired TrkB signaling in HD.