**DESCRIPTION FOR THE GENERAL PUBLIC (IN ENGLISH)**

The increase in ammonia concentration in the blood and/or brain tissue (hyperammonemia) is an accompanying factor of a *plethora* of metabolic disorders named hyperammonemic encephalopathies among which acute and/or chronic hepatic encephalopathy (HE) are the most common. Central nervous system (CNS) is particularly sensitive to the increased ammonia level. In the brain, astroglial cell are the main locus of ammonia detoxification by a direct glutamine synthesis *via* an enzymatic reaction catalyzed by glutamine synthetase. Simultaneously, under conditions of elevated ammonia level, impaired astrocytic function seems to be a primary cause of glutamatergic neurotransmission impairment observed in HE pathology. SN1(SNAT3; *Slc38a3*) is the major astrocytic glutamine transporter belonging to system N. SN1 is responsible for glutamine transport from astrocytes do adjacent neurons and as such contributes to the regulation of glutamatergic neurotransmission. The current view is that SN1 is regulated on transcriptional and translational level. So far, it was shown that SN1 activity in astrocytes is regulated by protein kinase C. What is more, recent literature data indicates that Sp1 transcription factor may play an important role in the regulation of SN1 expression. It was demonstrated that Sp1 upregulates SN1 expression in mouse kidney in metabolic acidosis. In the Department of Neurotoxicology, we have shown a decrease of SN1 mRNA expression and reduced SN1 protein level in frontal cortex of mouse brain with acute HE. Described effect was observed selectively in astrocytic-neuronal co-culture, but not in homogenous cultured primary cortical astrocytes. On the other hand, SN1 expression in ammonia-treated astrocytes appeared to be sensitive to Sp1 transcription factor silencing.

The literature data and our preliminary results prompted us to propose a project in which we will analyze the role of Sp1 transcription factor in the regulation of SN1 expression in ammonia-treated astrocytic cell cultures and astrocytic-neuronal co-cultures *in vitro* and brain slices of mouse with acute HE and hyperammonemia *ex vivo*. All proposed models are routinely used in our Department. Dependence of SN1 expression from the presence of Sp1 will be measured in real-time qPCR and Western Blot technique on astrocytes and astrocytic-neuronal co-cultures with silenced Sp1. Sp1 silencing will be obtained using siRNA sequences or Sp1 inhibitor. Analysis whether chosen Sp1 binding sites are enhancers or silencers of target gene will be performed by evaluation of luciferase expression in luminescence measurement. The interactions between SN1 and Sp1 binding sites will be investigated using chromatin immunoprecipitation (ChIP) method which uses properties of binding between the gene of interest (here: SN1) and sequences of transcription factor binding sites (here: Sp1). SN1 activity in the presence or absence of Sp1 transcription factor will be analyzed by measurement of labeled [$^{3}$H]Gln transport experiments in ammonia-treated astrocytes/co-cultures and mouse brain slices with induced HE/hyperammonemia.

Verification of the hypothesis that Sp1 transcription factor is involved SN1 expression in astrocytes may give an insight into a possible mechanism of SN1 regulation in the described pathological conditions. In addition, it may expand our knowledge about astrocyte-neuron interaction and its influence on impaired neurotransmission observed in HE pathology.