The role of sphingosine kinases/sphingosine-1-phosphate dependent signaling in mice brains exposed to type II diabetes mellitus and high-fat diet. Searching for potential neuroprotective effect of fingolimod and metformin.

Type 2 diabetes mellitus (T2DM) and obesity rapidly affects millions of people around the world, lowers their quality of life and have social and economic effects. The data from epidemiological studies demonstrate that, between 1980-2014 the number of adults with diabetes increased globally 4 times. Moreover, T2DM increases risk of other diseases, such as: cardiovascular disease, stroke, Alzheimer's disease (AD), finally leading to patients death. The etiologic basis of these most common metabolic disorders are still poorly understood.

High levels of ceramides are observed in obesity and this fact could be associated with T2DM. Ceramides has been intensively studied in relation to cell death induction and stress response. Recent literature concentrate on the role of ceramides in glucose homeostasis and insulin signaling. It also links high level of ceramides to insulin resistance - which is main pathological hallmark of T2DM. Moreover, it was observed that elevated ceramide levels may interrupt the existing balance between ceramide and pro-survival molecule - sphingosine-1-phosphate (S1P).

Until today, the role of sphingosine kinases/S1P (SphK/S1P) in brains of T2DM and in obesity induced by high-fat diet (HFD) remains unknown, so the following study will be conducted to evaluate this issue. **The fundamental goal of present project is to evaluate the role of S1P signaling in mice brains exposed to HFD and administration of low-doses of streptozotocin (STZ), which is accepted model for T2DM.** Concomitantly, the effect of HFD will be examined. Additionally, we plan to verify the neuroprotective effect of fingolimod (FTY720, which is a modulator the S1P receptors signaling) in T2DM and HFD mice models and to compare it with metformin (MET, which is a drug used to treat high-glucose levels in T2DM).

In this project the molecular/biochemical analysis of different parts of brain (cortex, hippocampus) will be performed to examine the alterations of proteins crucial in: S1P metabolism and action; protection against oxidative stress; inflammatory response as well as in glucose/insulin transport and signaling, amyloid precursor protein metabolism and Tau protein phosphorylation. In our research we will use following molecular/biochemical methods: gene expression analysis by Real-time RT-PCR, analysis of protein level by western blot (WB) and enzymes activity by chemiluminescent assays. Then, we will examine ultrastructural mitochondrial and synaptic terminals alterations in T2DM and HFD mice brain before and after fingolimod and metformin treatment. Finally, we will evaluate the cognitive function of T2DM and HFD animals by selected behavioral tests.

Diabetes represents an important clinical, economic and social problem. Rapidly increasing numbers of T2DM are not only associated with popularization of western (fast-food) diet, lack of physical activity and obesity but also with lack knowledge about the pathomechanism and poor control of the disease. The results of the following study will deliver novel information about the alterations of SphK/S1P metabolism and hopefully will help to understand the role of SphK/S1P signaling in pathomechanism of T2DM. Moreover, the molecular changes in SphK/S1P signaling in the brain of mice exposed to HFD will be evaluated. We assume that modulation of S1P signaling may reduce oxidative stress, protect against apoptosis and alter the metabolism pathways responsible for amyloid-β generation and Tau protein phosphorylation. We also expect that pharmacological treatment of T2DM mices with fingolimod will improve their cognitive functions.