Disrupted SorCS2-dependent neuronal protein sorting in neonatal epileptic encephalopathy: functional studies in human iPSC-based models.

Neonatal encephalopathy is brain damage caused by a reduction in oxygen partial pressure in the blood. One of the consequences of this phenomenon is neonatal epilepsy – a severe neurological disorder characterized by seizures, which can lead to cognitive, behavioral, and sensory impairments in newborns. Hypoxia can lead to cell damage, which in some individuals is associated with the development of epilepsy. The risk of developing epilepsy may be genetically determined. Genetic studies of children who have developed this disease have identified mutations in genes associated with epilepsy, which has sparked significant interest among scientists in the mechanisms underlying this condition.

As part of an international collaboration with scientists from the University of Alberta in Canada, we discovered a new mutation (c.2614C>T, p.Pro872Ser) in the *SORCS2* gene in a child suffering from neonatal epilepsy. The *SORCS2* gene encodes a neuronal receptor involved in intracellular transport. It is responsible for the recycling and sorting of membrane proteins, which is crucial for the functioning of synapses and brain plasticity. SorCS2 also affects the activity of glutamate receptors, which play an important role in signal transmission between neurons.

The aim of our project is to understand how the newly discovered Pro872Ser mutation affects neuronal function and what mechanisms underlie its association with epilepsy. These studies are important because they will help us better understand the processes related to the recycling of neuronal proteins and their role in the proper functioning of the brain. Disruptions in this process can lead to abnormal neuronal activity and contribute to the onset of epileptic seizures. In our project, we plan to conduct experiments on human neurons derived from induced pluripotent stem cells (iPSC). Using organoid models, three-dimensional structures resembling miniature brains, it will be possible to study the effect of the mutation at the tissue level as well. By analyzing neuronal development and activity, we will be able to determine how disruptions in protein sorting and recycling affect their function. Our research will also test how cells with the Pro872Ser mutation respond to standard antiepileptic drugs. Additionally, we will analyze a variant of cells lacking the *SORCS2* gene (SorCS2 KO) to better understand the impact of its absence on neuronal function and epilepsy-related processes in human neurons. This could provide valuable information about the effectiveness of current therapies and assist in developing new treatment strategies. We will use a wide range of research methods, including live neuron imaging, proteomic analyses, RNA sequencing, and electrophysiological measurements of neuronal activity.

We expect that our results will contribute to a better understanding of the mechanisms underlying epilepsy and identify potential therapeutic targets for treating children suffering from genetically determined forms of this disease.