Among animal cells neuronal cells (neurons) have the most complicated morphology and functions. The complex morphology of neurons reflects the role they play in analyzing information by the nervous system. These cells with help of their extensive cytoplasmic protrusions (dendrites and axons) form a complex connection network, in which dendrites receive signals from other members of the network while axon transmits information to the subsequent circuit elements. The generation of nerve cells, including acquisition of the correct neuronal shape is known as neurogenesis. This process is regulated by a number of molecular mechanisms, some of which engage protein called mTOR. mTOR integrates information about resources cell has at its disposal (e.g., availability of amino acids, or energy level) with extracellular growth stimulating signals. If, mTOR "decides" that the internal resources allow the cell growth, by means of its target proteins, initiates necessary molecular changes needed for example for neuroprogenitor migration and differentiation or dendritic growth of developing neurons.

The key molecular mechanism required for movement and growth of all kinds of cells including neurons is correct transport of a variety of cellular elements along cellular highways - microtubules. Transportation of any cargo along microtubules requires the involvement of proteins called molecular motors: dynein-dynactin complex and kinesin. Often one type of motor has capability to carry multiple types of cargo. The specificity of cargo binding to motors is provided by another group of proteins named adaptors. So far, there was no data showing directly mTOR involvement in the regulation of the activity of motors and adaptors. However, our recent preliminary data point to a possibility that mTOR alters the binding of some adaptors to dynactin.

The main goal of this project is to test the hypothesis that during neuronal development mTOR modulates the dynamics of motor-adaptor interaction thus contributing to the adjustment of intracellular transport to the needs of growing cell. At the same time, we plan to check whether disturbance of the process may be involved in the development of tuberous sclerosis, a disease arising from excessive activity of mTOR, resulting in several neurodevelopmental symptoms.

The proposed research will be conducted on several levels of complexity. At the beginning using biochemical methods, electron and light microscopy we plan to describe in detail how mTOR modulates interaction between motor and adaptor using as a role model new motor-adaptor pair which we recently discovered, namely dynactin and β-adaptin. In addition, we will check whether the same model of interaction modulation by mTOR is generally applicable for binding of other adaptors to dynactin. Then employing the combination of available cellular (neuroblastoma cells, primary cultures of neurons, neural stem cells obtained by means of somatic cells reprogramming) and in vivo (e.g., the developing zebrafish brain) models with ultra-fast confocal microscopy, we will test if mTOR is indeed needed for long-distance journeys of selected cargo along microtubules in nerve cells. Moreover, we will verify if mTOR-dependent modulation of adaptor-motor binding affects selected aspects of the nervous system development (e.g. the formation of dendritic trees) under physiological conditions and those mimicking tuberous sclerosis. In the final part of the project we plan to test hypothesis that mTOR may also affect the function of kinesin - a second group of molecular motors.

As a result of this research project we expect to describe a new mechanism of regulation of intracellular transport in nerve cells undergoing intensive growth during neuronal network formation. In addition, we will provide information on the potential contribution of molecular motors to the pathogenesis of tuberous sclerosis, which will contribute to a better understanding of the molecular basis of this still mysterious disease.