Ischemia of brain and heart tissue is the one of the most common causes of death worldwide. Recent studies have revealed that mitochondria play the crucial role in cellular death. Basic function of mitochondria, called cellular powerplants, is energy production which used in many processes in the cell. It has been found that ischemia induces mitochondrial damage what promotes cell death. Discovery of natural cytorpotective mechanisms preserving mitochondrial function during ischemia is goal of many studies. Recent years mitochondrial potassium have been identified. These proteins regulate potassium fluxes accross the mitochondrial inner membrane. The basic properties have benn found to be similar to the potassium channels from the plasma membrane. It has been found that activation of mitochondrial potassium channels with pharmacological substances preserves mitochondria against damage induced with various factors including ischemia. The mechanism of this phenomenon remains unclear.

Additionally, it has been found that in the cell small amounts of gases (nitric oxide, carbon monoxide and hydrogen sulfide) are synthesized. These substances regulate many proteins including some of the potassium channels. Studies have revealed that these molecules might regulate activity of potassium channels from plasma membrane inducing cytoprotective mechanisms. However it is not clear whether activity of the mitochondrial potassium channels is regulated by these gaseous signaling molecules. The present project aims to describe the molecular mechanism of regulation of mitochondrial potassium channels by gaseous signaling molecules (NO, H_2S and CO) and functional consequences of this process.

Presented research will focus on regulation of two channels: mitochondrial large conductance potassium channel regulated by calcium (mitoBK_{Ca}) and mitochondrial potassium channel regulated by ATP (mitoK_{ATP}). Project will be divided into three parts.

First, we will focus on the describing the effects induced by gasotransmitters on the activity of the mitochondrial potassium channels. In our research we will use two cell lines as experimental models. Activity of the mitoBK_{Ca} channel will be studied using astrocytoma U87-MG cells. The second experimental model will be a cardiac H9c2 cell line. From both cell lines we will isolate mitochondria, which will be used for electrophysiological studies. Our laboratory is equipped with unique systems for the patch-clamping of mitoplasts (swollen mitochondria). This experimental approach allows for monitoring of single channel activities in isolated mitochondria. The biggest advantage of this unique methodology is based on the fact that studied channels are in their native enviroment. To modulate activity of the mitochondrial potassium channel it is planned to use chemical donors of the gasotransmitters and known pharmacological modulators of the channel.

The second aim of the project will focus on answering question regarding the mechanism of mitoK_{ATP} channel regulation by NO, H_2S and CO. This will be possible because recently it has been published that a protein called ROMK2 is the main component of the channel. We plan to develop several cell lines expressing mutated ROMK2 protein. Point mutations will be introduced in the positions of the potential binding sites for gases. In the last part of the project we will induce injury of the cells using oxidative stress and hypoxia. We will measure cell death of clones expressing mutated ROMK2 protein and compare it with cells expressing wild type of the channel. Previous studies have revealed that overexpression of the wild type ROMK2 protein protects cells against cell death induced by oxidative stress.

Results of the planned research will help to understand natural cellular mechanisms of cytoprotection. It is possible, that obtained data will provide hints for development of new therapeutic strategies against consequences of damage induced by ischemia of heart and brain tissue.