Tumor diseases are one of the most common cause of death on the word. One of the main hallmark of tumor is rapid progression of abnormal cells that may spread to other organs in process termed metastases.

Due to the fact that available antitumor treatment involving chemotherapy, radiotherapy and surgery are insufficient estimates are staggering - prevalence of tumor may increase to 19.3 million per year by 2050. All kinds of treatment cause side effects, since they are characterized by nonspecific action in addition to normal and tumor cells. Tumorigenesis is the multistep process closely associated with significant alterations on molecular level within cell. They are caused by stress stimuli such as glucose deficiency, disturbances in redox homeostasis and level of calcium.The major stress stimuli which leads to rapid tumor progression is hypoxia, which elicits cellular response pathways which are closely connected with alterations in gene expression.

We predict that utilising small molecular PERK inhibitors may contribute to overcame current problems associated with ineffectiveness of therapy against tumor diseases. Under hypoxic condition tumor cells, in comparison with normal cells, significantly slowdown processes required oxygen and energy consumption. The source of this difference lies in adaptive processes of tumor cells to survive under hypoxic stress. Low oxygen level induces endoplasmic reticulum (ER) stress, which leads to activation one of the transmembrane endoplasmic reticulum (ER) receptor PERK. As a result the Unfolded Protein Response (UPR) pathways are activated. There is a lot of evidence that UPR signalling cascade have a dual role. On the one hand ensues phosphorylation one of the translation initiation factor eIF2α. This significant for tumor cells modification causes downregulation of global protein synthesis. Moreover, it also leads to cell cycle arrest in growth phase (G1). In turn, under prolonged stress conditions expression of two specific protein such as ATF4 and CHOP are upregulated. Crucial for our research is the fact that they are closely affiliated with activation ER-dependent apoptotic pathways leading to programmed death of tumor cells. There is very little research that examine this paradox of the UPR in tumor diseases. The newest data suggest that there is a possible correlation between two variables: high level of CHOP protein and low of another cytoplasmic protein p21 which also may potentially a dual pro-apoptotic and pro-survival role.

We hypothesize that utilising small molecule PERK inhibitors switch from pro-adaptative to pro-apoptotic signalling pathways in stress condition. We have already initiated proposed project. Due to the fact that that we currently conducting different project in collaboration with the Medical University of South Carolina we were able to select set of new potential PERK inhibitors utilizing docking software and HTS screen. Nine compounds, with the highest PERK specificity were selected from 79552 compounds, which we would like to test in proposed project. We plan to conduct our research on five kinds of tumor cell lines such as: A549, BxPC3, HT29, SH-SY5Y, IMR32, that will be treated with thapsigargin, as cellular model of endoplasmic reticulum-dependent stress and UPR response.

Testing our hypothesis involves five main research tasks:

1. Assessment of biological activity of PERK inhibitors by evaluating phosphorylation of eIF2α using the anti-phospho-eIF2α (S51) specific antibody in Western Blot. We will use cell lines treated with thapsigargin and as a positive control PERK knockout cells ($PERK^{-/-}$),
2. Analysis of cytotoxic effect of selected PERK inhibitors,
3. Analysis of PERK/eIF2α/ATF4 signalling activation under normal or hypoxia conditions. QPCR and Western Blot (WB) will be used to assess the expression and phosphorylation level of PERK and eIF2α as well as its downstream dependent proteins CHOP and ATF4 under normal or hypoxia conditions,
4. Analysis of possible crosstalk between PERK/eIF2α/ATF4 pathway and p21 by manipulating the expression of p21,
5. Analysis of the level of apoptosis and cell cycle progression by DeadEnd™ Fluorometric TUNEL System in cells under normoxia or hypoxia with or without PERK inhibitor treatment,

The proposed project is among the first which can give the answer how to overcame the drawbacks of current antitumor therapies. We predict that utilising small molecule PERK inhibitors as a potentially anti-neoplastic therapeutics may provide effective elimination of tumor cells and as a result contribute to development novel, targeted antitumor strategy and release many people form death.