Synthetic analogues of opioid growth factor linked to derivatives of glucosamine as a compounds with the potential for targeted therapy against pancreatic tumour

The pancreatic cancer is the fourth leading cause of cancer-related death, despite the fact it is relatively rare type of cancer. This is due to one of the lowest 5-year survival rate, which is 6.9% (average for Europe). More than 80% of patients are identified with locally advanced and metastatic stage. According to the previous studies, OGF (met-enkephalin) has been shown inhibitory effect on the cancer growth, including pancreatic cancer on *in vitro* as well *as in vivo* model. The aim of the project is search of new substances with potential clinical application in pancreatic cancer treatment. The research includes synthesis of analogues of opioid growth factor (OGF): met-enkephalin, with established anticancer activity. Moreover, met-enkephalin showed a beneficial clinical effect in patients with pancreatic adenocarcinoma by inhibited tumour growth simultaneously did not exhibit significant side effects. The synthesis of conjugates with glucosamine derivatives is aimed at enhance anticancer activity through established immunostimulat activity of glucosamine. In the first step of biological study, by use MTT and LDH assay, it will be assessed effect of the obtained anaolgues on the cells viability: pancreatic cancer cells compare with immortalized hTERT-HPNE and primary renal mixed epithelial cells. Compounds exhibited the best cytotoxic profile will be used to evaluate their antiproliferative activity by the BrdU incorporation assay. In order to determine non-specific cytotoxic and cytostatic activity of synthesized analogous in PANC-1, MIA PaCa2 and Capan-1, OGFR gene will be knok-out. Also, the impact on cell cycle by flow cytometry, the level of proteins associated with regulation of cell cycle and programmed cell death: autophagy, apoptosis and necroptosis will be investigated. Analysis of structural and ultrastructural changes in pancreatic cancer cells treated with selected complexes, will be performed by fluorescence microscope and transmission electron microscope. Because met-enkephalin exhibits antiangiogenic activity, it will be performed the tube formation assay on human HUVEC cell line. Also the effect of test compounds on platelet activation by pancreatic cancer cells using unique equipment - QCM-D, imitating the conditions prevailing in the human microvascular system will be determined. From healthy volunteers will be collected blood and centrifuged to obtain peripheral blood mononuclear cells. Subsequently, in separated blood lymphocytes and monocytes the immunostimulatory activity of synthesized analogues will be performed by detection cytokines releasing into the supernatant. The next step of the biological study will be included primary pancreatic cancer cells that will be surgically removed from patients in the university clinic. In the last step of our project, after toxicological and pharmacokinetic study, we will evaluate anticancer activity of selected compounds on transgenic mice with pancreatic cancer. The obtained results will be evaluated by statistical methods using GraphPad PRISM. The result of the project will provide the basis for further extended *in vivo* study, which ultimately could lead to patent application and bring new drug to treat pancreatic cancer.