

Chasing the HARE: elucidation of the binding mechanism of the new and unexplored ligands of the Hyaluronic Acid Receptor for Endocytosis, stabilin-2.

The aim of this project is to determine the three-dimensional structure of functional domains of the Hyaluronic Acid Receptor for Endocytosis (HARE, or stabilin-2) by means of X-ray crystallography, as well as to characterize the binding of the chosen new ligands to this receptor.

The project constitutes basic research in life sciences, in particular, in the field of biochemistry, molecular biology and structural biology.

The main reason for conducting the research on stabilin-2 is its considerable involvement in many physiological processes and pathologic states (e.g. cancer progression) and in drug delivery to the target cells, as well as its connection with diet and nutrition.

Stabilin-2 belongs to the group of the scavenger receptors and plays a crucial role in clearance of more than 10 ligands from bloodstream, including products of degradation of the extracellular matrix and metabolic products. One of these ligands is hyaluronic acid that binds to the Link domain of stabilin-2. Present knowledge of ligands recognition and binding, as well as mechanisms of signal transmission by stabilin-2, is fragmentary and incomplete.

It has recently been demonstrated that the stabilin-2 knock-out or blocking of the receptor by an antibody effectively opposes cancer metastasis by elevating the level of the circulating hyaluronic acid. Moreover, due to the fact that stabilin-2 is also responsible for the binding and internalization of advanced glycation end products, it may be also involved in the processes connected with e.g. diabetes, another important civilization disease.

Recent research has shown that stabilin-2 is entangled also in many processes connected with drug delivery into the target cell. It recognizes e.g. oligonucleotides (short DNA fragments), which may find a use as medicines regulating the process of expression of unwanted proteins. The investigated HARE receptor leads to endocytosis and internalization of the oligonucleotides, which in this way enter the target cells. Stabilin-2 also binds and introduces to the cell the nanoparticles coupled to hyaluronic acid and potential drug, which may be as well an effective strategy for the drug delivery. Mechanisms of interaction of stabilin-2 with these important ligands are not well explained. Due to the high structural similarity, it is not excluded that stabilin-2 may also bind RNA oligonucleotides.

Only one of stabilin-2 domain has been described until today - the FAS1 domain (published by the Principal Investigator). Determination of the three-dimensional structure of the Link domain of stabilin-2, responsible for hyaluronic acid and other ligands binding (possibly with the contribution of neighbouring domains), would provide valuable information for understanding the mechanism of the ligand binding and clearance. Determination of the crystal structure of stabilin-2 would also allow for the rational design of the tightly binding small-molecule antagonists of this protein and, consequently, open up a prospect for new chemical probes for studying the signaling in the stabilin-2 pathway. Another reason for the proposed research is that the structural information can enhance structural studies on proteins with a similar domain organization and function. Moreover, the detailed biochemical characteristics of the hyaluronan binding by HARE would bring more insights into the involvement of this complex in cancer progression and metastasis.