Thermodynamic contribution of a halogen bond in protein-ligand systems. Design, synthesis, and thermodynamic studies of model systems.

Past years' events have shown us how vital in pharmacology is to optimize the research and rapidly screen large amounts of biologically active compounds in terms of their interaction with the particular molecular target. One of the steps to be taken in the drug design process is the thermodynamic analysis of interactions in the system formed by the target protein and biologically active substances (ligands). We determine the complete set of thermodynamic parameters connected with a given system. There is a tendency to obtain the most energetically favourable form in nature. This phenomenon can be used as an engine to gain desired effect in any biological system.

Free Energy of Gibbs (ΔG) is an energy change associated with any reaction. Based on this value, we can predict how the system would behave. For example, while the ΔG value is negative, we can say that reaction occurs spontaneously. Thus the determination of ΔG is crucial to making a detailed thermodynamic description of the studied system. We must determine the energetic contribution of all interactions occurring between protein and its ligand to do that. With the use of computer techniques, we can currently predict the contribution of most types of interactions. An exception is halogen bonds, the type of interactions occurring between a halogen atom and proximal electron-rich objects, whose contribution to ΔG is estimated from 0.2 to 7 kcal/mol. For the precise determination of binding free energy, such discrepancy is unacceptable. In this project, we will try to deliver information that should help determine the thermodynamic contribution of halogen bonds in protein-ligand systems

The catalytic subunit of casein kinase CK2 interacting with its inhibitors – halogenated heterocyclic compounds, will be used as the research model. Casein kinase is the enzyme responsible for regulating metabolic pathways involved in cell divisions and apoptosis (controlled suicide of cell). Increased CK2 level has been found in many types of cancers. This phenomenon is probably responsible for accelerated divisions and the high survivability of cancer cells. In this way, inhibition of this enzyme became of interest to pharmacology. Halogenated benzotriazole derivatives are competitive inhibitors of CK2. Because of the simplicity of structure with four possible halogen atoms, they are perfect probes for our research.

Numerous interactions between ligand and the protein were identified in crystallographic structures obtained for CK2 and brominated benzotriazole derivatives. Besides halogen bonds and salt bridges, the multiple positioning of ligands in the active centre was also visible. The influence of hydrophobicity was also evident while introducing bromine atoms to the structure causes them to escape from the aqueous environment to the hydrophobic part of the protein. With so many variables, it is impossible to determine the contribution of a halogen bond to ΔG. This project will try to decrease the number of variables by using specially designed ligands and the protein. We are using variant α’ of the catalytic subunit of CK2 (hCK2α’), which, according to crystal structures, does not show multiple positioning of the ligands inside the active centre. Specially synthesized compounds, which are equally hydrophobic and differ only in halogenation patterns, will eliminate the variation of hydrophobicity and electronic properties. Crystal structures delivered by collaborators from the University of Cologne indicated, that compounds chosen by us bind to hCK2α’ in the same way, despite different halogenation patterns. With the determined free energy of ligand binding for such complexes, with only one variable describing the pattern of halogen bonds, we will be able to estimate the contribution of a single halogen bond to ΔG. Thus we will make the thermodynamic description of a biological system much more complete.