“Structure, function and dynamics: How biological macromolecules and related tools help to understand ligand-protein and protein-protein interactions”

Abstract:
Research of our group is primarily focused on peptide chemistry and biology as well as protein biochemistry using methods of bioorganic synthesis and bioanalytics. Using two examples, i.e. heme-protein (1) and peptide-protein (2) interactions, the usefulness of peptides as models as well as bioactive compounds in this respect will be demonstrated.

(1) The progress in heme-related research is intriguing and still broadening our knowledge about the function of this small molecule. During the last two decades the capability of heme to act as a signaling molecule was shown. Heme has crucial regulatory effects in vital functions of the organism such as transcription, metabolism, ion channel opening, and in the immune system. In addition, it is supposed to be one of the key players in diseases such as Alzheimer’s, cancer, and stroke/cerebral vasospasm. In the last years the understanding of heme binding to heme-regulated proteins at distinct sites on the protein's surface was improved based on a combinatorial peptide library approach and a set of peptides representing characteristics for specific heme-binding. However, there is still an apparent lack of knowledge regarding the mode of interaction, i.e. the molecular basis for heme association to a particular protein. In a collaborative project the knowledge on protein regulation by free heme is increased with the aim to address new challenging issues such as the prediction of so-far unknown heme-regulated proteins or the identification and structural characterization of heme interaction sites on relevant proteins.

(2) Cysteine-rich peptides are widespread and possess different functions in nature that are in part related to their conserved tertiary structure. In our laboratory we focus on oligopeptides containing more than four cysteine residues, e.g. different conotoxins from marine cone snails, which possess interesting features concerning their bioactivity and a huge potential for pharmaceutical and medicinal applications. A previous work of our lab reported the isolation of different disulfide-linked isomers of µ-conotoxin PIIIA. These different disulfide isomers have been identified using 2D-NMR technique, however, another method to elucidate the disulfide linkage is MS/MS sequencing of digested and derivatized peptide fragments. In addition, another Cys-rich peptide, the 66mer tridegin, which is a useful inhibitor of factor XIIIa of the blood coagulation cascade derived from the giant amazon leech, was focus of our interest, too. Interestingly, MS/MS analysis of this peptide revealed a completely different disulfide connectivity compared to the previous mentioned conotoxins and, in addition, during oxidation three different isomers were formed sharing one out of three disulfide bonds. Thus, LC-ESI MS/MS is a powerful tool for the elucidation of disulfide connectivities of disulfide-rich peptides in connection with NMR structural analysis, however, requires carefully optimized protocols for individual peptide representatives.