Report for 3rd Quarter (February - April 2005)

TArget Specific Scoring FUNctions (TASSFUN)

The TASSFUN project, supervised by Dr. Niklas Blomberg (GSI CompChem AZ Mölndal) and Dr. Rebecca Wade (EML Research, Heidelberg), aims at the construction of target specific tailor-made scoring functions by protein structure-based COMparative BINding Energy (COMBINE) analysis. The method provides the possibility to derive 3D QSARs for a set of receptor-ligand complexes whose 3D structures can be modeled. As target proteins, trypsin-like serine proteases of the blood coagulation cascade were selected because of the large amount of published data as well as their importance in diseases. Among these proteases, we have so far focused mainly on the urokinase-type plasminogen activator (uPA).

In the first six months, we worked on developing programs for semi-automatic preparation of x-ray structures for molecular mechanics calculations and also on acquisition of respective published kinetic and structural data in SDF files. Initial COMBINE models for uPA and trypsin were built with a training set of ligands with a common structural element of a 2-(2-phenol)indole or a 2-(2-phenol)benzimidazole moiety. During the third quarter of the TASSFUN project, we have adapted the programs written at EML Research to the AZ CCE-P/Protease Platform during a visit in Mölndal from February 19th to 25th and improved them to run the step of structure preparation in a more automatic way in batch jobs suitable for large amounts of data. Simultaneously, problems of parameterisation of ligands were solved by using SDF files for specifying bond orders in ANTECHAMBER. Finding the right protonation state for the receptor-ligand-complexes is still a difficult task. At the moment, we add hydrogens first to the ligand with InsightII, and then to the protein and ligand with WHATIF.

The COMBINE model of uPA was extended to all 36 published x-ray structures with 29 different ligands. Based on these structures and the corresponding electron density maps, six models of uPA with flexible residues in various alternative conformations were built. These models should be used for flexible docking of ligands of unknown binding conformations. To validate the docking procedure, we have started to dock ligands back into the crystal structure of the uPA receptor and compared the docked binding mode with the experimental one. The docking calculations have shown a requirement of some water molecules for obtaining the correct binding mode. For this reason, all 36 crystal structures were superimposed and nine potentially important water sites were manually selected. Which water sites are important for the docking procedure and perhaps, also for COMBINE models will be checked in further docking calculations. As soon as we have specified the required water sites and have a suitable docking protocol, we will start virtual screening with uPA models and small molecule test sets of binders and non-binders.