SIXTH FRAMEWORK PROGRAMME

Protein Interactions with the Environment

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Interactions of proteins with their environment are fundamental for understanding biological and hybrid systems consisting of biological and inorganic compounds. The environment of proteins is composed of solvent, other proteins and surfaces.

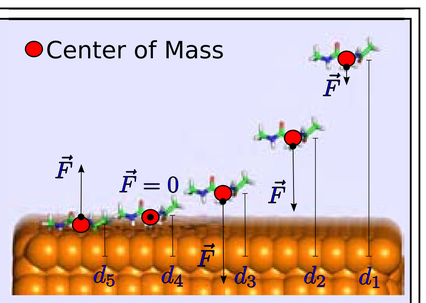
Methods

Molecular Dynamics

Our reasearch has been conducted with the **Gromacs** Molecular Dynamics package. As Force Field, **OPLS-AA** (Optimized Potential for Liquid Simulations) has been chosen and extended for our aminoacid - gold simulations.

Potential of Mean Force Calculation

The PMF along a chosen **reaction coordinate** allows the calculation of the **Free Energy difference** between two states. The method of choice in our systems is the evaluation of **constraint forces** (see ref. 1&2) in simulations with constrained distances on the **reaction coordinate**. In case of the Barnase - Barstar model system, we chose the **COM - COM** (Center of Mass) distance as our reaction coordinate while the **COM distance** of the aminoacid **from the topmost gold layer** plane has been the choice in our gold systems. 21 distances (27 in the gold systems) have been sampled with at least 4x5ns simulations. The obtained mean force profiles are integrated to their potential form.



References: 1. Trzesniak, D.; Kunz, A. P. E. & van Gunsteren, W. F.; *A comparison of methods to compute the potential of mean force* ; ChemPhysChem, 2007 , 8 , 162-169, 2. Mulders, T.; Kruger, P.; Swegat, W. & Schlitter, J.; *Free energy as the potential of mean constraint force* ; Journal Of Chemical Physics, 1996 , 104 , 4869-4870

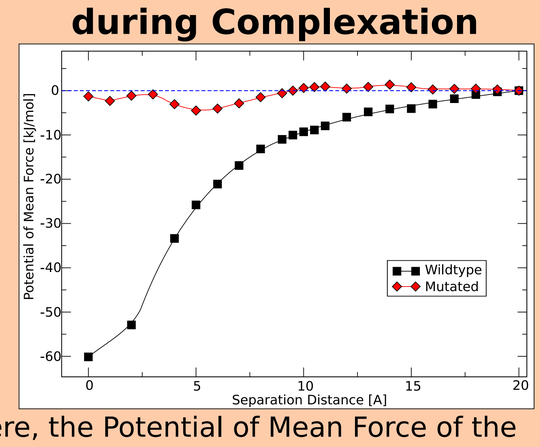
Complexation of Transient Protein Complexes: The Barnase-Barstar Model System

Motivation

Protein-protein interactions are essential for many cellular processes. For transient complexes, **electrostatic steering** has an important contribution to the **association of proteins**. This contribution depends on the distribution of charges across the complexation partners as well as on the properties of the **surrounding solvent**. Contrary to macroscopic systems, the solvent properties are not homogeneous and isotropic but therefore depend on the surrounding **protein surfaces**.

We studied the well known system consisting of a ribonuclease (Barnase) and its inhibitor (Barstar). To analyze the **impact of mutations** on the electrostatic steering, we mutated Lys27 and Arg 59 on Barnase as well as Asp39 and Glu76 on Barstar to Alanine. Simulations at various **constrained Center of Mass** (COM) distances were conducted while monitoring the **constraint force** and orientation of the water molecules during the simulation.

Potential of Mean Force during Complexation



Here, the Potential of Mean Force of the Barnase - Barstar wildtype complex and a mutant with strongly reduced electrostatic interaction is shown along the reaction coordinate. The distance is the additional separation from complex COM-COM distance in the wildtype crystal structure (1BGS).

Water Dipoles during Complexation Process

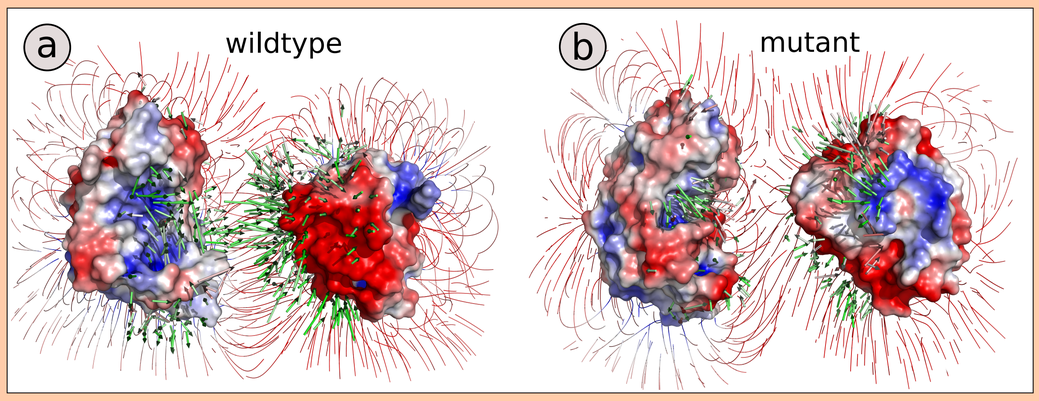
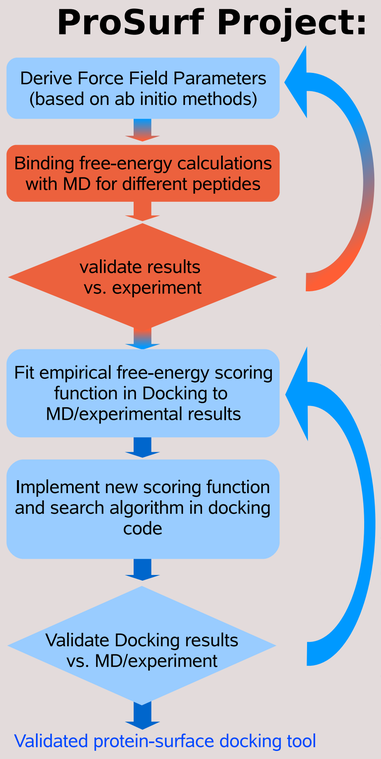


Figure a) and b) show the complexation of Barnase and Barstar at constrained separation distance of 20A. The electrostatic field is shown as fieldlines and the water dipoles are displayed as arrows. While the mutant is still connected with a field of prealigned water molecules prealigned water is present only at specific surface positions in the mutant complex. Induced water structure trough prealignment can reduce the dielectric constant ϵ and further improve protein complexation via electrostatics.

MD Simulations of Aminoacid Interactions with 111 oriented Gold Surfaces

ProSurf Project: Motivation and Workflow

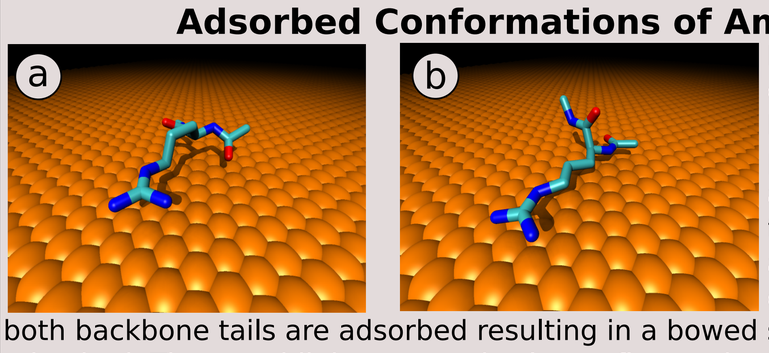


The focus of group LMU is marked in red

In biological circumstances, proteins interact not only with their counterpart but also with **inorganic surfaces** like bone. **Compatibility** with non-biological surfaces and classification of protein - surface interactions is of increasing **importance for nanotechnology** and **drug design**. Yet, a physical under-standing for these interactions is currently lacking.

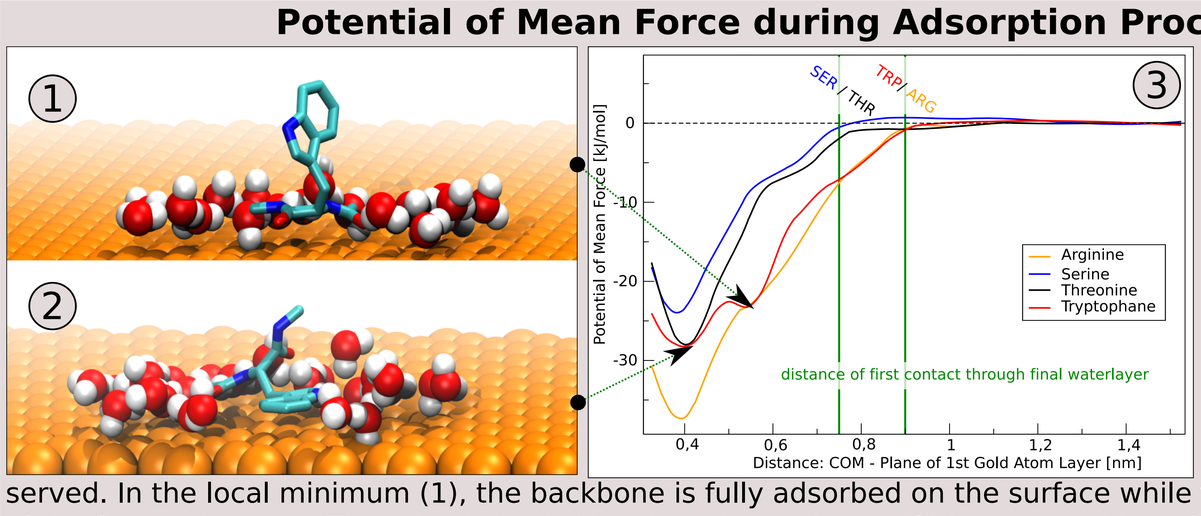
The major target of the **ProSurf** project is the **development of a toolkit** allowing the characterization of protein - surface interactions. In this task we evaluate the derived **force field parameters** (INFM-CNR Modena) and verify the **docking** results (EML Heidelberg). The **experimental verification** of the **parameterization process** is achieved in cooperation with Nanoscience and Biochemistry groups (Weizmann Institute of Science).

Adsorbed Conformations of Aminoacids on 111 Gold Surfaces



Free simulations allow the investigation of adsorbed conformations. This is of importance in two aspects: First, we can verify the impact of parameters by comparing adsorbed conformations with those obtained via ab-initio (DFT) calculations. Second it allows us a structural insight in the adsorption and potential cooperative effects. Figure a) and b) show conformations of capped aminoacids adsorbed on the gold surface. In a) both backbone tails are adsorbed resulting in a bowed sidechain tail while in b) only one backbone tail is adsorbed. The guanidinium group is always flat on the gold surface.

Potential of Mean Force during Adsorption Process

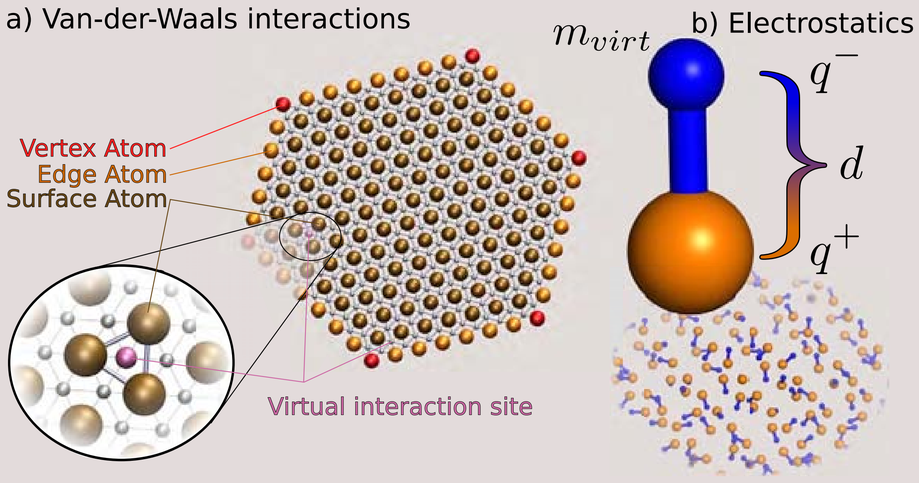


PMF from different capped aminoacids. The distance of first contact of trough the final waterlayer coincides with the first significant decrease of potential during adsorbtion. In the PMF trace of Tryptophane (3), two distinct minima in the Free Energy can be observed. In the local minimum (1), the backbone is fully adsorbed on the surface while the sidechain sticks out of the final waterlayer. The adsorbtion in the absolute minimum (2) is governed by a flat adsorbed conformation of the aromatic sidechain together with one of the two backbone tails adsorbed.

Parameterization of Gold Surfaces

a) Van-der-Waals interactions

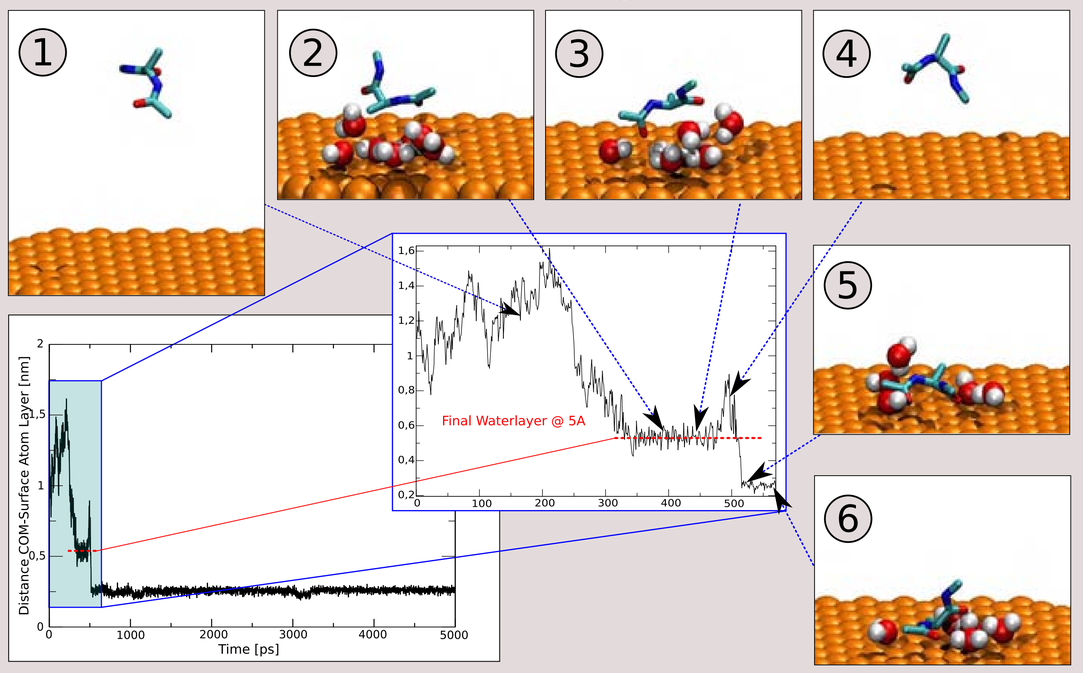
b) Electrostatics



a) and b) illustrate the implementation of interactions with the 111 oriented gold surface. The van-der-Waals interaction (ϵ,σ) is carried by virtual interaction sites in the geometric center of each triangle formed by surface gold atoms as shown in a). Electrostatic interactions are modeled with dipoles at the position of gold atoms shown in b). During simulation, the virtual interaction sites and charge on the gold atom position is frozen while and the virtual mass $m_{virt}$and its charge is chained to the gold atom with a distance constraint.

Dewetting Transition of Aminoacids Adsorbing on 111 Gold

To adsorb on the gold surface, the final separating waterlayer as a barrier needs to be overcome. Here a typical dewetting process of a capped aminoacid (Alanine) is shown in the time-distance trace with corresponding simulation snapshots. The barrier at a distance of 5A in the time-distance trace is clearly visible in frame (2) and (3) as the final waterlayer. The adsorbtion process in free simulations starting from 1nm distance is very fast for the uncharged peptides (~500ps). No desorption events could be observed from any peptide during 5ns simulation time.



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