AKB 90.4 Mi 14:45 TU H2013

How parallel is protein (un)folding? — •Lothar Reich and Thomas R. Weikl — Max Planck Institute of Colloids and Interfaces, Theory Division, 14424 Potsdam

According to the 'old view', proteins fold along well-defined *sequential* pathways, whereas the 'new view' sees protein folding as a highly *parallel* stochastic process on funnel-shaped energy landscapes. We have analyzed parallel and sequential processes on a large number of Molecular Dynamics unfolding trajectories for the protein CI2 at high temperatures. Using rigorous statistical measures, we find that the degree of sequentiality depends on the structural level under consideration. On a coarse substructural level of whole β-sheets and helices, unfolding is predominantly sequential. In contrast, the unfolding process is more parallel on the level of individual contacts between the residues of the protein chain. On an intermediate structural level, the characteristic parallel and sequential events can be understood from simple loop-closure dependencies between the substructural elements.

AKB 90.5 Mi 15:00 TU H2013

All Atom Protein Structure Prediction with Stochastic Optimization Methods — •Wolfgang Wenzel, Thomas Herges, Alexander Schug, and Abhinav Verma — Forschungszentrum Karlsruhe, Institut für Nanotechnologie, Postfach 3640, 76021 Karlsruhe

The prediction of protein tertiary structure remains one of the outstanding problems in biophysical chemistry. According to the thermodynamic hypothesis, the native conformation of a protein can be predicted as the global optimum of its free energy surface with stochastic optimization methods[1] orders of magnitude faster than by direct simulation of the folding process.

We have recently developed an all-atom free energy forcefield (PFF01)[2] which implements a minimal thermodynamic model based on physical interactions. With this forcefield we were able to predictively fold the 20 amino acid trp-cage protein[3], the 40 amino-acid HIV accessory protein[4], the 36 amino-acid villin headpiece and the 60 amino acid bacterial ribosomal protein[5] using various stochastic optimization methods. We will disccuss advantages and limitations of these methods with respect to further improvements of this approach to in-silico all-atom protein strcuture prediction.

[1] W. Wenzel, K. Hamacher, PRL 59, 3003 (1999) [2] T. Herges, W. Wenzel, Biophys. J. 87, 3100 (2004) [3] A. Schug, W. Wenzel, PRL 91, 158102, 2003, EPL 67, 307 (2004) [4] T. Herges, W. Wenzel, PRL (in press) [5] A. Schug, W. Wenzel, JACS (in press)

AKB 90.6 Mi 15:15 TU H2013

Identification of Oxygen Channels in Proteins by Molecular Dynamics — •Jan Saam, Christopher Ozdoba und Hermann-Georg Holzhütter — Institut für Biochemie, Charité, Monbijoustr 2. 10117 Berlin

Cells contain a variety of enzymes that use molecular oxygen in the reactions they catalyze. In most cases the influence of oxygen-protein interaction on the reaction is unknown. We employed molecular dynamics simulations to determine the oxygen pathway from the solvent phase to the active site and to study the oxygen adsorption at the inner surface of two different oxygenases.

Our results show that in each enzyme there exists an oxygen channel different from the substrate entrance leading through the protein matrix to the catalytic site. The channels cannot be seen in the crystal structure but open their different segments temporarily yet allowing oxygen molecules to diffuse to the active center. With its high probability density for oxygen the interior end of the tunnel represents the ideal point for the stereo- and position specific insertion of dioxygen into the substrate. Subsequently these results could be confirmed by mutation experiments.

AKB 100 Poster Session I

Zeit: Samstag 16:45–18:45 Raum: Poster TU D

AKB 100.1 Sa 16:45 Poster TU D

Activation and characterization of a photoswitchabel GFP variant using two-photon absorption — •Marc Schneider[1], Sara Barozzi[2], Ilaria Testa[1], Mario Faretta[2], and Alberto Diaspro[1] — [1]INFM Genua, Via Dodecaneso 33, I-16146 Genoa, Italy — [2]European Institute of Oncology, Dept of Exp. Oncology, Via Ripamonti 435, I-20141 Milan, Italy

We report about a photoactivatable variant of the Aequorea Victoria green fluorescent protein (PA-GFP). As reported by Patterson and *Lippincott − Schwartz*[1] this special form of the molecule increases its fluorescence intensity when excited by 488 nm after irradiation with high intensity light with $\lambda = 413nm$. We will present data on the two-photon induced photoactivation of the PA-GFP molecule as well as two-photon excitation. Therefore experiments were performed using partially purified protein immobilised on microspheres. The molecular switches were irradiated with laser light in a range of wavelength of a Ti:Sapphire laser system coupled to an inverted microscope. The optimum frequency for activation was chosen to investigate fixed cells. A comparison between the conventional activation with a single photon at $\lambda = 405nm$ and two-photons demonstrates the much smaller activation volumes in the cell.
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(1) Patterson, G. H.; Lippincott-Schwarz, J. Science 2002, 297, 1873.

AKB 100.2 Sa 16:45 Poster TU D

Thermal Fluctuations of Individual Semiflexible Polymers in Confined Geometry — •Sarah F. Köster[1,2], Stephan Herminghaus[1,2], Myung C. Choi[3], Cyrus R. Safinya[3] und Thomas Pfohl[1,2] — [1]Department of Applied Physics, University of Ulm, Albert-Einstein-Allee 11, 89081 Ulm, Germany — [2]Max Planck Institute for Flow Research, Bunsenstraße 10, 37073 Göttingen, Germany — [3]Materials Research Laboratory, University of California, Santa Barbara, CA 93106, USA

Thermal fluctuations of individual actin filaments in confining microchannels fabricated by soft photolithography are studied by means of fluorescence microscopy. The channel dimensions are in the same order of magnitude as the mesh size of the actin cytoskeleton within the eukaryotic cell and thus mimic the native environment of the individual filament. We observe a strong dependence of the tangent correlation upon both the channel geometry and the filament length. Compared to freely fluctuating filaments, long filaments confined in narrow channels exhibit an enhanced tangent correlation revealing a local minimum and an oscillatory behavior. We also observe a clear deviation from existing theoretical models on small length scales, assumedly due to an intrinsic stiffness of the semiflexible chain. These unique characteristics may be qualitatively described by an analytical expression considering the bending energy as well as the confining energy assumed as a parabolic potential. We find the scaling law for the deflection length which has been reported before experimentally confirmed.

AKB 100.3 Sa 16:45 Poster TU D

Interactions of the Extracellular Matrix Protein Collagen I and the Actin Cytoskeleton — •Sarah F. Köster[1,2], Jennie B. Leach[2], Joyce W. Wong[2] und Thomas Pfohl[1] — [1]Max Planck Institute for Flow Research, Bunsenstraße 10, 37073 Göttingen, Germany — [2]Department of Biomedical Engineering, Boston University, 44 Cummington Street, Boston, MA 02215, USA

Both the extracellular matrix (ECM) where collagen is the most important building block and the actin cytoskeleton impact the mechanical properties of mammalian tissue. The study of these fibrous proteins and all the more their interaction is thus a very interesting field whenever looking at living beings. We use a microfluidic diffusive mixing device to create a defined pH gradient in a microchannel which in turn initiates the polymerization and concurrent alignment of soluble collagen into fibrils under hydrodynamic flow. We are thus able to investigate collagen fibrillogeneisis by means of polarization microscopy and x-ray diffraction. Furthermore, substrates prepared by using this technique are used as scaffolds for cell growth. Since the collagen structure has precise alignment in native blood vessels, study of the impact of highly anisotropic (aligned) collagen on vascular smooth muscle cells (VSMC) provides much-needed insights towards structure-property-function relationships between the ECM and the cytoskeleton. Anisotropic collagen induces alignment of the cytoskeleton and may facilitate the study of the cytoskeleton by means fluorescence microscopy and in addition by x-ray diffraction.