BP 56: Protein Structure and Dynamics

Time: Thursday 15:00–17:30 Location: SCH A251

Invited Talk BP 56.1 Thu 15:00 SCH A251

Biophysical Studies of Amyloid Formation and Its Inhibition — •Sheena Radford — The University of Leeds

Amyloid formation involves the polymerisation of proteins and peptides into polymers with a cross-beta fold. How amyloid formation causes disease, and identifying the culprit species involved, remain a significant challenge. This results from the complexity of the aggregation process and the fact that amyloid assembly is initiated by non-native states of proteins that are partially folded or intrinsically disordered. Structure determination is thus difficult, and identifying the interacting surfaces in these transiently formed and dynamic ensembles is challenging. In this lecture I will describe our recent attempts to discover new insights into how proteins aggregate into amyloid and how to prevent cellular dysfunction caused by amyloid assembly/disassembly mechanisms using a number of different strategies. Specifically, I will show how we have used different biophysical and biochemical approaches to map the nature of the earliest protein-protein interactions in amyloid assembly and to re-assess the potential role(s) of fibrils in disease. Finally, using a novel screen developed with E.coli, we have been able to discover new highly potent inhibitors of aggregation for some of the most highly aggregating protein sequences known.

BP 56.2 Thu 15:30 SCH A251

Molecular mechanisms of substrate binding proteins in ABC transporters — Marijn de Boer, Florence Husada, Kostas Tassis, Yusran Muthahari, Giorgos Gouridis, and •Thorben Cordes — Zernike Institute for Advanced Materials, University of Groningen, Nijenborgh 4, 9747AG Groningen, The Netherlands

ATP-binding cassette (ABC) transporters mediate unidirectional active transport of diverse substrates across membranes using ATP hydrolysis. During import in prokaryotes, specialized substrate binding proteins or domains (SBDs) at first capture a substrate for delivery to the translocator domain and subsequent transport. Different SBDs show a high structural conservation but only little sequence similarity. Their fold consists of two rigid α/β domains, which form the binding cavity, and that are connected by a (flexible) hinge. We here test the hypothesis, whether there is a relationship between the SBD hinge structure and conformational dynamics of the SBDs based on the fact that the hinge bends during the conformational transition from open unliganded to closed liganded state. For this we use single-molecule Förster resonance energy transfer (smFRET) that allows to elucidate the conformational states and dynamics of SBDs directly and with this to understand their mechanistic role for transport.[1-3]

[1] G. Gouridis et al., Nature Structural & Molecular Biology 22 (2015) 57-64. [2] F. Husada et al., Biochemical Society Transactions 43 (2015) 1041-1047. [3] J. H. M. van der Velde et al., Nature Communications 7:10144 (2016).

BP 56.3 Thu 15:45 SCH A251

Time-resolved observation of allosteric communication in PDZ2 domain — •Gerhard Stock, Sebastian Buchenberg, and Florian Sittel — Institute of Physics, Albert Ludwigs University, Freiburg

A local perturbation of a protein may lead to functional changes at some distal site. A well-established example are PDZ domains which show an allosteric transition upon binding to a peptide ligand. Recently Hamm and coworkers (PNAS 2013) presented a time-resolved study of this transition. Using well-defined photopreparation and structure-sensitive infrared probing, they showed that the conformational rearrangement of PDZ2 occurs in a highly nonexponential manner on various timescales from pico- to microseconds.

Here we present extensive (in total 0.4 ms) nonequilibrium molecular dynamics simulations (all atom/explicit solvent) of Hamm's experiment, which allow us to monitor protein allosteric communication in real time. Along these lines, we address the following issues: What are the timescales of the transition and is there a hierarchy of processes? Is allosteric communication in PDZ2 dominated by enthalpic or entropic effects? Can the process be described as directed motion (domino picture), a simple barrier crossing, or rather via a diffusive heterogeneous transition path ensemble? Is allostery connected to the protein's pathways of energy transport? What's the role of the solvent? Does the long-range conformational rearrangement due to ligand binding occur via population-shift or induced-fit mechanism?

BP 56.4 Thu 16:00 SCH A251

Elucidation of light-induced structural changes of aureochrome by small-angle X-ray scattering — •Saskia Bannister, Elena Herman, Thomas Hellweg, and Tilman Kottke — Bielefeld University, Germany

Aureochromes function as blue-light-regulated transcription factors in algae. Their basic region leucine zipper (bZIP) effector domain binds DNA with a specific sequence while a light-, oxygen-, or voltage-sensitive (LOV) domain acts as the C-terminal sensor. Due to this unusual arrangement of sensor and effector, aureochromes are interesting for studying their mechanism and for the engineering of new synthetic optogenetic tools.

We are applying small-angle X-ray scattering (SAXS) to resolve the solution structure of the full-length receptor and the monomeric LOV domain. However, SAXS on photoreceptors is challenging. First, dark conditions need to be absolutely strict to avoid conversion of the highly sensitive receptor. Second, the analysis under illumination needs to ensure full conversion. Therefore we are establishing SAXS experiments under rigorous control of light and simultaneous UV/Vis monitoring on an in-house setup. First results from SAXS measurements on LOV under true dark conditions are presented. Experiments on the full-length aureochrome are currently pursued.

Banerjee, A., Herman, E., Serif, M., Maestre-Reyna, M., Hepp, S., Pokorny, R., Kroth, P. G., Essen, L.-O., Kottke, T. (2016), *Nucleic Acids Res.* 44(12), 5957-5970.

Herman, E., Kottke, T. (2015), *Biochemistry* 54, 1484-1492.

BP 56.5 Thu 16:15 SCH A251

Barrier crossing in the presence of long memory - a global analysis — •Julian Kappler, Jan O. Daldrop, Florian N. Brünig, and Roland R. Netz — Freie Universität Berlin, Germany

Rate theories are a widely used and important means for predicting meso- and macroscopic time scales from microscopic dynamics. The present rate theories for non-Markovian microscopic dynamics, i.e. in the presence of memory effects, are quite complicated to implement and have not been studied systematically for dynamics with long memory time. In our contribution, we present a global analysis of barrier crossing rates based on simulations of the massive Langevin equation with exponential memory. We extract the scaling behavior of the rates for long memory, indicate limits of current rate theories, and additionally provide a heuristic formula to calculate accurate rates quickly and easily.

BP 56.6 Thu 16:30 SCH A251

Color tuning of visual rhodopsins: a quantitative explanation by electrostatic calculations — •Florimond Collette, Frank Müh, and Thomas Renger — Institut für Theoretische Physik, Johannes Kepler University Linz, Altenberger Strasse 69, 4040 Linz, Austria

Rhodopsins are biological pigment-protein complexes found in photoreceptor cells of the retina. Comparing the results of two quantum chemical/electrostatic calculation methods, that have been applied successfully to reveal the functional states of BLUF photoreceptors [1] and photosystem II antenna protein CP29 [2], we have estimated absorption shifts of the retinal chromophore for a series of site-directed mutants. Our results are in excellent agreement with recent experimental studies [3] and strongly suggest that the spectral sensitivity in animal rhodopsins is dominated by electrostatic tuning.

[1] F. Collette et al., *J. Phys. Chem. B* **118**, 11109 (2014).
[2] F. Müh et al., *Phys. Chem. Chem. Phys.* **16**, 11848 (2014).
[3] W. Wang et al., *Science* **338**, 1340 (2012).

BP 56.7 Thu 16:45 SCH A251

The dynamics and flexibility of penicillin binding proteins: a combined computational/experimental approach to tackle antimicrobial resistance (AMR) — •Jasmine L. Desmond[1], Pierdomenico Bellini[2], Christopher Dowson[2], P. Mark Rodger[3], and Rudo A. Roemer[1] — [1]Department of Physics,

University of Warwick, UK. — [2]School of Life Sciences, University of Warwick, UK. — [3]Department of Chemistry, University of Warwick, UK.

700,000 people die each year from drug-resistant infections, a figure that — if action is not taken — is estimated to increase to 10 million by 2050. The drug penicillin targets essential cell wall biosynthetic enzymes that still remain attractive targets for new efforts in drug discovery. Elucidating protein dynamics and flexibility is key to understanding the selective interactions of proteins with a drug as it docks. In spite of the success of x-ray crystallography in the determination of rigid protein structures, the experimental technique is unable to provide insight into the dynamics of proteins. Such information can, however, be elucidated using molecular modelling. Important protein conformational changes often occur on microsecond-millisecond timescales and are difficult to access using traditional modelling techniques, such as molecular dynamics (MD). Here, we present the results of computationally inexpensive, geometric simulations of protein motion for a range of penicillin binding proteins. There is a focus on differences in motion between: (1) inactive and active proteins and (2) proteins with and without bound drug molecules.

BP 56.8 Thu 17:00 SCH A251

Femtosecond Time-Resolved Dynamics in Myoglobin observed with an XFEL — •Henrike M. Müller-Werkmeister[1,2], Helen M. Ginn[3], Anling Kuo[4], Antoine Sarracini[2], Helen Duyvesteyn[3], Sascha W. Epp[1], Darren Sherrell[5], Shigeki Owada[6], Olivier Pare-Labrosse[2], Saeed Oghbaey[2], Jessica Besaw[2], Yoshiaki Kumagai[7], Kensuke Tono[6], Yinpeng Zhong[1], Koji Motomura[7], Bryan T. Eger[2], Alexander Marx[1], Arwen R. Pearson[8], Robin L. Owen[5], Kiyoshi Ueda[7], David I. Stuart[3,5], Oliver P. Ernst[4], and R. J. Dwayne Miller[1,2] — [1]Max-Planck-Institute for Structure and Dynamics of Matter, Hamburg, Germany — [2]Departments of Chemistry and Physics, University of Toronto, Canada — [3]University of Oxford, United Kingdom — [4]Department of Biochemistry, University of Toronto, Canada — [5]Diamond Light Source, Didcot, United Kingdom — [6]SACLA, RIKEN Spring-8 Center, Hyogo, Japan — [7]Tohoku University, Sendai, Japan — [8]Hamburg University, The Hamburg Center for Ultrafast Imaging, Germany

Here we present recent results from femtosecond time-resolved serial crystallography experiments performed at the X-Ray free electron laser SACLA. We have studied the ligand dissociation in Myoglobin in the time window between 0 fs and 2 ps under functionally relevant conditions, that is, at low laser excitation conditions to prevent spurious artefacts introduced by multi photon processes. The results provide an unprecedented insight into the ultrafast structural changes right after the ligand dissociation.

BP 56.9 Thu 17:15 SCH A251

A rigid core amplifies the binding affinity of multivalent ligands — •Susanne Liese and Roland R. Netz — Department of Physics, Free University Berlin, Arnimallee 14, 14195 Berlin, Germany

Multivalent interaction achieves strong, yet reversible binding through the simultaneous formation of multiple, identical bonds. The strength of multivalent binding is determined by the interplay between gain in binding enthalpy and entropic loss due to the reduction of conformational and rotational degrees of freedom upon ligand binding. We compare the binding affinities of rigid and flexible multivalent ligands. While flexible polymer based scaffolds are unsuitable to design high affinity multivalent ligands, rigid ligands with a stiff core that is similar in size to the receptor, amplify the binding affinity by several orders of magnitude. The general design principles, which we derive, might be of great importance to improve rational multivalent ligand design.