# BP 3: Protein Structure and Dynamics

Time: Monday 9:30–12:15 Location: H44

**Invited Talk** BP 3.1 Mon 9:30 H44

**Structural Dynamics of Single G-protein Coupled Receptors** — •Emmanuel Margeat — Centre de Biochimie Structurale, UMR 5048 CNRS, INSERM U1054, Université de Montpellier, France

Efficient cell-to-cell communication relies on the accurate signaling of cell surface receptors. Understanding the molecular bases of their activation requires the characterization of the dynamic equilibrium between active and resting states. Here, we monitor, using single molecule Förster resonance energy transfer, the kinetics of the activation of metabotropic glutamate receptor (mGluR), a class C G-protein coupled receptor (GPCR) activated by glutamate, the major excitatory neurotransmitter in the central nervous system. By combining filtered Fluorescence Correlation Spectroscopy (fFCS), excited stated lifetime analysis and Photon Distribution (PDA) analysis on single diffusing receptors, we demonstrate that most receptors oscillate between a resting- and an active- conformation on a sub-millisecond timescale. Interestingly, we demonstrate that differences in agonist efficacies stem from differing abilities to shift the conformational equilibrium toward the fully active state, rather than from the stabilization of alternative static conformations, which further highlights the dynamic nature of mGluRs and revises our understanding of receptor activation and allosteric modulation.

BP 3.2 Mon 10:00 H44

**Scaling rules for vibrational energy transport in globular proteins** — •Sebastian Buchenberg geb. Waltz[1], David M. Leitner[2], and Gerhard Stock[1] — [1]Biomolekulare Dynamik Physik Uni-Freiburg — [2]Chemistry University of Nevada/Reno

Computational studies of vibrational energy flow in biomolecules have to date mapped out transport pathways on a case by case basis [1]. To provide a more general approach, we derive scaling rules for vibrational energy transport in a globular protein, which are identified from extensive nonequilibrium molecular dynamics simulations of vibrational energy flow in the villin headpiece subdomain HP36 [2]. We parameterize a master equation based on inter-residue, residue-solvent and heater-residue energy transfer rates which closely reproduces the results of the all-atom simulations. From that fit two scaling rules emerge. The first for the energy transport along the protein backbone which is described by a diffusion model in which the local diffusion strongly depends on the size of the individual amino acid side chain. And the second for the energy transport between tertiary contacts which is based on a harmonic description and depends on the coupling strength of the contact and the charge of the atoms in contact. Requiring only the calculation of mean and variance of relatively few atomic distances, the approach is able to predict the pathways and timescales of vibrational energy flow in proteins.

[1] D. M. Leitner, S. Buchenberg, P. Brettl and G. Stock, J. Chem. Phys. 142, 075101 (2015) [2] S. Buchenberg, D. M. Leitner and G. Stock, submitted (2015)

BP 3.3 Mon 10:15 H44

**Peptides in Presence of Aqueous Ionic Liquids: Tunable Co-Solutes as Denaturants or Protectants?** — Volker Lesch[1], Andreas Heuer[1], Diddo Diddens[1], Christian Holm[2], and •Jens Smiatek[2] — [1]Institut für Physikalische Chemie, Westfälische Wilhelms-Universität Münster, D-48149 Münster, Germany — [2]Institut für Computerphysik, Universität Stuttgart, D-70569 Stuttgart, Germany

We studied the stability of a small beta-hairpin peptide under the influence of aqueous 1-ethyl-3-methylimidazolium acetate ([EMIM]+ [ACE]-) solution via all-atom molecular dynamics simulations in combination with metadynamics. Our free energy results indicate a denaturation of the peptide structure in presence of the ionic liquid which is validated by a significant broadening of the end-to-end distance. The radial distribution functions between the ions and the peptide were used for the calculation of the preferential binding coefficients in terms of the Kirkwood-Buff theory. A significant structure dependent binding of acetate to the peptide was found which can be interpreted as the main reason for the denaturation of the native conformation. The outcomes of our simulations allow us to propose a simple mechanism to explain the unfolding of the peptide with regard to the specific properties of ionic liquids. Our results are in good agreement with experimental findings and demonstrate the benefits of ionic liquids as tunable co-solutes with regard to their influence on protein structural properties.

BP 3.4 Mon 10:30 H44

**The C-terminus of human copper importer, Ctr1, acts as binding site and transfers copper to Atox1** — •Michael Kovermann[1,2], Dana Kahra[2], and Pernilla Wittung-Stafshede[2,3] — [1]Fachbereich Chemie, Universität Konstanz, Germany — [2]Department of Chemistry, Umeå University, Sweden — [3]Department of Biology and Bioengineering, Chalmers University of Technology, Sweden

Uptake of copper ions (Cu) into human cells is mediated by the plasma membrane protein Ctr1, followed by Cu transfer to cytoplasmic Cu chaperones for delivery to Cu-dependent enzymes. The C-terminal cytoplasmic tail of Ctr1 is a 13-residue peptide harboring a HCH motif thought to interact with Cu. We here employ biophysical experiments under anaerobic conditions to peptide models of the Ctr1 C-terminus to deduce Cu-binding residues, Cu affinity and ability to release Cu to the cytoplasmic Cu chaperone Atox1. Based on NMR assignments and bicinchoninic acid competition experiments, we demonstrate that Cu interacts in an one-to-one stoichiometry with the HCH motif with an affinity $K_D$ of $10^{-14}$ M. Removing either the Cys residue or the two His residues lowers the Cu-peptide affinity but site specificity is retained. The C-terminal peptide and Atox1 does not interact in solution in the absence of Cu. However, as directly demonstrated at the residue level via NMR spectroscopy, Atox1 readily acquires Cu from the Cu-loaded peptide. We propose that Cu binding to the Ctr1 C-terminal tail regulates Cu transport into the cytoplasm such that the metal ion is only released to high-affinity Cu chaperones.

**30 min break**

BP 3.5 Mon 11:15 H44

**Dynamics of dissolved BSA studied by QENS: MD simulations compared to experiments** — •Christian Beck[1], Marco Grimaldo[2], Felix Roosen-Runge[2], Tilo Seydel[2], Fajun Zhang[1], and Frank Schreiber[1] — [1]Institut für Angewandte Physik, Universität Tübingen, 72076 Tübingen — [2]Institut Laue - Langevin, Grenoble, France

Recent improvements in the field of quasi-elastic neutron scattering (QENS) offer the possibility to explore simultaneously the global movements and the internal dynamics of proteins in solution [1,2]. While the global diffusion can be described by colloidal models, different non-consistent descriptions exist for the internal dynamics. To address this challenge, scattering functions are calculated from atomically resolved molecular dynamics (MD) simulations of bovine serum albumin in solution for a temperature series crossing the denaturation temperature, analysed with a two-state model of switching diffusing processes and compared with experimental data [1].Furthermore, we implemented the model of fractional Brownian dynamics [3].
For both models, the analysis is expanded onto broader energy windows compared to the experimental one to test the limits of the models and alsoto open the possibility to combine results from different neutron spectrometers such as time-of-flight and backscattering instruments.

[1] M.Grimaldo et al. Phys. Chem. Chem. Phys., **17** (2015) 4645
[2] M.Grimaldo et al. J. Phys. Chem. B **118** (2014) 7203
[3] I.Krasnov et al. Phys. Rev. E **91** (2015) 042716

BP 3.6 Mon 11:30 H44

**Molecular dynamics study of the mechanical stability of dimeric coiled-coils under strain** — •Chuanfu Luo, Ana Vila Verde, and Reinhard Lipowsky — Max Planck Institute of Colloids and Interfaces, 14476 Potsdam, Germany

Coiled-coils are ubiquitous folding motifs found in proteins. They consist of two alpha-helices wrapped around each other in a super helix configuration. In biological systems, coiled coils are highly versatile: they play an important role in various intracellular regulation processes as well as in membrane fusion. Their unusual structure suggests that it may also be possible to use them as biological force sensors to detect forces involved in biochemical processes in vivo. We investigated this

possibility by carrying out Steered Molecular Dynamics to simulate the shear pulling of de novo designed coiled-coils with different lengths. We find that the pulling force at slow constant pulling speed is independent of either the total length or the contact length of the coiled-coils, and is the same as the force for unfolding a single alpha-helix. The results suggest that short coiled-coils under slow shear strain move via a dislocation mechanism: a defect is created at the pulled end of the helix and travels to the other end of the helix in tens of nanoseconds.

BP 3.7 Mon 11:45 H44

**Photo-dynamics of photoactivated adenylyl cyclase LiPAC from the spirochete bacterium *Leptonema illini* strain 3055** — •Alfons Penzkofer[1], Meenakshi Tanwar[2], Sindu Kandoth Veetil[2], and Suneel Kateriya[2,3] — [1]Fakultät für Physik, Universität Regensburg, Universitätsstraße 31, D-93053 Regensburg, Germany — [2]Department of Biochemistry, University of Delhi South Campus, Benito Juarez Road, New Delhi 110021, India — [3]School of Biotechnology, JNU, New Delhi 110067, India

The photoactivated adenylyl cyclase LiPAC from the spirochete bacterium *Leptonema illini* was synthesized and characterized by absorption and fluorescence spectroscopic methods [1]. LiPAC consists of a BLUF domain and an adenylyl cyclase homology domain. Photo-excitation of fully oxidized flavin in LiPAC resulted in a typical primary BLUF domain photo-cycle dynamics. The quantum efficiency of BLUF domain signaling state formation was determined to be 0.60. Continued blue-light-excitation of LiPAC in the light-adapted state caused irreversible photo-degradation of non-covalently bound flavin to covalently bound fully reduced flavin with a quantum efficiency of $1.1\times10^{-5}$. At 20 °C the time constant of signaling state recovery to the receptor state after excitation light switch-off was 2.6 s. The protein thermal stability was studied by stepwise sample heating and cooling. A LiPAC melting temperature of 54 °C was determined. Schemes of the primary BLUF domain photo-cycling dynamics and the secondary BLUF domain photo-degradation in the signaling state are presented.

[1] A. Penzkofer et al., Trends in Applied Spectroscopy 11 (2014) 39.

BP 3.8 Mon 12:00 H44

**Protein anisotropy modulates the coupling between rotational and translational diffusion under crowding conditions** — •Matthias Roos[1], Maria Ott[1], Marius Hofmann[2], Susanne Link[1], Jochen Balbach[1], Ernst Rössler[2], Alexey Krushelnitsky[1], and Kay Saalwächter[1] — [1]Martin-Luther-Universität Halle-Wittenberg, Institut für Physik, Germany — [2]Universität Bayreuth, Lehrstuhl Experimentalphysik II, Germany

*In vivo* molecular motion of biopolymers is known to be strongly influenced by excluded-volume effects caused by the high concentration of organic matter inside cells, usually referred to as crowding conditions. In order to further understand the effects on translational and rotational diffusion, we performed pulsed-field gradient and field-cycling NMR, X-ray scattering and viscosity measurements for three proteins in water solution - $\alpha$B-crystallin, bovine serum albumin and lysozyme. Our results demonstrate, on the one hand, that long-time translational diffusion quantitatively follows the expected increase of macro-viscosity upon increasing the protein concentration. The behavior of rotational diffusion, on the other hand, turns out to be protein-specific and spans the full range between the limiting cases of full coupling and full decoupling from the macro-viscosity. We show that the anisotropy of inter-protein interactions, in particular of electrostatic nature, is the main factor modulating the (de)coupling between rotational and long-time translational diffusion.