

BP 7: Posters: Biological Machines, Motor Proteins

Time: Monday 17:15–20:00

Location: Poster B1

BP 7.1 Mon 17:15 Poster B1

Modeling on the hydrodynamic interaction between microswimmers — ●JOHANNES GREBER and RUDOLF FRIEDRICH — Institut für Theoretische Physik, WWU Münster, Wilhelm-Klemm-Str. 9, 48149 Münster

We are interested in swimming bacteria consisting of a head and a couple of flagella. Generally it shows two states of motion: On the one hand by bundling the counterclockwise rotating flagella the bacterium translates. On the other hand the bacterium tumbles or rotates, when one flagellum rotates clockwise. Using the velocity field created in the surrounding fluid by these movements bacteria can interact with each other, which leads to the question, if there are collective effects among several bacteria.

Assuming that the velocity field of one bacterium consists of two rigidly coupled point vortices in some given distance, we derive the equations of motion for the case of interaction between two swimming objects. Moreover, we make some predictions for the trajectories of the swimmers with the help of a linear stability analysis.

BP 7.2 Mon 17:15 Poster B1

Monitoring a single Sec translocase complex in an anti-Brownian electrokinetic (ABEL) trap — ●TORSTEN RENDLER¹, STEFAN ERNST¹, KARIN SEYFERT¹, ANDREAS KUHN², and MICHAEL BÖRSCH¹ — ¹3. Physikalisches Institut, Pfaffenwaldring 57, 70569 Stuttgart, Germany — ²Institut für Mikrobiologie und Molekularbiologie, Universität Hohenheim, Germany

Translocation of polypeptides in *E. coli* cells is catalysed by the membrane protein complex SecAYEG. This process is powered by ATP (adenosine triphosphate) hydrolysis of the SecA motor component. To investigate the translocation process, SecAYEG is reconstituted into lipid vesicles and the conformational changes during polypeptide transport are monitored by internal fluorescence resonance energy transfer (FRET). Therefore, the different SecAYEG subunits were labeled with various fluorescent markers. Previous confocal measurements of single translocases in solution suffered from the limited observation time due to Brownian motion. To increase the observation time we combined a confocal setup for FRET measurements with an anti-Brownian electrokinetic (ABEL) trap. The ABEL-trap was developed by A.E. Cohen (Harvard) and W.E. Moerner (Stanford) and is based on an active feedback mechanism consisting of a EMCCD camera to locate the complex and electrodes to apply an electrical field across the trapping region. We present preliminary FRET data of a single translocases held in solution by the ABEL-trap.

BP 7.3 Mon 17:15 Poster B1

Cooperative effects in the inhibition of a Kinesin-5-head/Kinesin-1-stalk chimera by monastrol — STEFAN LAKÄMPER^{1,2,3}, CHRISTINA THIEDE¹, ●ANDRÉ DÜSELDER¹, STEFANIE REITER^{1,3}, LUKAS C. KAPITEIN^{2,4}, ERWIN J.G. PETERMAN², and CHRISTOPH F. SCHMIDT^{1,2,3} — ¹Drittes Physikalisches Institut, Georg-August-Universität Göttingen, Germany — ²Department of Physics and Astronomy and Laser Centre, VU University Amsterdam, The Netherlands — ³DFG-Research Centre for Molecular Physiology of the Brain, CMPB, Göttingen, Germany — ⁴Current address: Erasmus University Medical Centre, Rotterdam, The Netherlands

Several kinesin motors are required for proper assembly of the mitotic spindle. The homo-tetrameric bipolar Kinesin-5 can cross-link and slide antiparallel microtubules apart by a motility mechanism comprising diffusional and directional motility. In order to explore the basic kinesin-5 motor activity, we generated a stably dimeric Kinesin-5 construct, Eg5Kin, consisting of motor domain and neck-linker of *Xenopus laevis* Kinesin-5 and neck coiled-coil of *Drosophila melanogaster* Kinesin-1. This chimera is highly processive. We studied the effect of the Kinesin-5-specific inhibitor monastrol in single-molecule fluorescence assays. In order to find out if one or two monastrol molecules terminate a run, we analyzed the monastrol concentration dependence of the motor run length. We found a Hill coefficient of about 2. We discuss in how far this means that two monastrols need to be bound to create an effect and what kind of cooperativity this implies for binding of monastrol to the two heads of a motor dimer.

BP 7.4 Mon 17:15 Poster B1

An in vivo approach to probing mechanotransduction apparatus function — ●BJÖRN NADROWSKI, THOMAS EFFERTZ, and MARTIN GÖPFERT — Abt. Zelluläre Neurobiologie, Universität Göttingen, MPI für Experimentelle Medizin, Hermann-Rein-Str. 3, 37075 Göttingen

The opening and closing of ion channels are mechanical events. These gating movements can be monitored in mechanosensitive ion channels provided that these channels are directly gated by force via macroscopic structures that thereby reflect the movements of the channels' gates. When coupled to molecular adaptation motors, these mechanosensitive ion channels form a transduction machinery that allows for active amplification while translating mechanical into electrical stimuli. Profiting from this experimental advantage, we have probed ion channel mechanics inside an intact *Drosophila* mechanosensory system. A physical model of this system is presented that quantitatively links ion channel mechanics, movements of molecular adaptation motors, and macroscopic mechanical events. Using this model, molecular parameters such as the gating energy required to open a single ion channel can be deduced. These energies have been determined for two force-gated ion channels. We also present evidence that these two channels are arranged in parallel in the transduction apparatus and may serve the detection of different stimuli amplitudes.

BP 7.5 Mon 17:15 Poster B1

A fast tetrameric Kinesin-5/Kinesin-1 chimera - a tool to study mechanisms of Kinesin-5 regulation — ●CHRISTINA THIEDE^{1,2}, STEFAN LAKÄMPER^{1,2}, ALOK D. WESSEL¹, STEFANIE REITER¹, and CHRISTOPH F. SCHMIDT¹ — ¹Drittes Physikalisches Institut, Georg-August-Universität Göttingen, Germany — ²these authors contributed equally to this work

The homo-tetrameric Kinesin-5 motor protein Eg5 from *X. laevis* drives relative sliding of anti-parallel microtubules (MT) by the processive action of its two opposing sets of dimeric motors. On a single MT, individual tetrameric motors move slowly (≈ 20 nm/s), but processively, alternating between a diffusional and a directional mode, while motors moving between two MTs move in a highly directional and processive fashion. In order to obtain a tetrameric model system with more easy discernable properties and motile phases, we have constructed a tetrameric chimera by replacing Eg5 motor domain and neck linker by the homologous regions of *D. melanogaster* Kinesin-1 (DK4mer). In surface-gliding assays, Dk4mer showed fast motility (553 ± 31 nm/s). Single GFP-tagged DK4mer motors moved processively along MT at comparable speeds (499 ± 3 nm/s). We observe clearly distinguished directional and diffusional episodes and an overall run length of $\approx 9 \mu\text{m}$. The DK4mer is thus an excellent model system to study regulatory aspects of Kinesin-5 due to its high speed, its long processivity and its clear separation of diffusive and directional motility.

BP 7.6 Mon 17:15 Poster B1

Direct observation of the myosin-V power stroke and its reversal — ●JAMES R. SELLERS³ and CLAUDIA VEIGEL^{1,2} — ¹Abteilung Zelluläre Physiologie, Institut fuer Physiologie, Ludwig Maximilians Universitaet Muenchen, Pettenkoferstrasse 12, Muenchen, Germany — ²Physical Biochemistry, National Institute for Medical Research, The Ridgeway Mill Hill, London NW7 1AA, UK — ³Laboratory of Molecular Physiology, National Heart, Lung and Blood Institute, NIH, Bethesda, MD USA 20892

Complex forms of cellular motility, including cell division, organelle trafficking or signal amplification in the auditory system, require strong coordination of the myosin motors involved. The most basic mechanism of coordination is direct mechanical interactions of individual motors that modifies their mechano-chemical cycles. Here, we used an optical tweezers-based single molecule assay to investigate the reversibility of the force generating conformational change (power stroke) of single myosin-V motor heads. By applying load to the head shortly after binding to actin, we found that at a certain load, the power stroke could be reversed. At this load the motor fluctuated between an actin-bound pre- and a post-power stroke conformation. This dramatic, load-dependent mechanical instability of a single motor head might be critical to coordinate the heads of processive, dimeric myosin-V. Interestingly, highly non-linear response to load, such as power stroke

reversal, can lead to coordination, synchronisation or even oscillations already amongst motors alone. These phenomena are critical for many cellular functions.

BP 7.7 Mon 17:15 Poster B1

A tetrameric Kinesin-1/Kinesin-5 chimera promotes fast relative sliding of microtubules — ●ALOK D. WESSEL, CHRISTINA THIEDE, STEFAN LAKAMPER, STEFANIE REITER, and CHRISTOPH F. SCHMIDT — Drittes Physikalisches Institut, Georg-August-Universität Göttingen, Germany

The Eg5 protein from *Xenopus laevis* is a homo-tetrameric motor protein which moves on microtubules (MT) in a processive manner and is capable of sliding two MT apart. Single motors show a directional motility with low velocity ($\approx 20\text{nm/s}$) as well as a diffusive behavior on MTs. The balance between directional and diffusive behavior is altered by cargo binding, i.e. crosslinking of MTs, or by a change of the ionic strength. In order to obtain a tetrameric model system with more clearly defined properties and motile phases, we constructed a chimera, DK4mer, by replacing Eg5-motor domain and neck-linker by the homologous regions of Kinesin-1 (*D. melanogaster*). Here we show that this tetramer, just like Eg5, promotes relative sliding when binding between two microtubules. DK4mer, however, slides two antiparallel MTs apart with a ~ 40 fold higher velocity than Eg5, between 700 and 1100nm/s. In multi-motor relative gliding assays, different binding geometries and velocities could be observed depending on the relative MT polarity and on residual motors on the substrate surface. We further measured the dependence of relative sliding on the ionic strength. Whereas surface gliding velocity remained unchanged, the relative velocity of two MT increased by $\sim 400\text{nm/s}$ when increasing salt concentration from 0 to 70 mM KCl in 30 mM Pipes buffer.

BP 7.8 Mon 17:15 Poster B1

Functional Rotation of the Transporter AcrB: The Essentials of Peristaltic Motion and Subsequent Substrate Extrusion — ●ROBERT SCHULZ¹, ATTILIO VITTORIO VARGIU², MICHAEL SCHREIBER³, PAOLO RUGGERONE², and ULRICH KLEINEKATHÖFER¹ — ¹School of Engineering and Science, Jacobs University Bremen, Germany — ²SLACS & Department of Physics, University of Cagliari, Italy — ³Institut für Physik, Technische Universität Chemnitz, Germany

The RND transporter of *E. coli*'s multidrug efflux pump AcrAB-TolC is able to export structurally and chemically different, toxic substrates, including antibiotics, via a functional rotation. The three major states of this rotation cycle were found in several asymmetric crystal structures. After initially analyzing the basic mechanisms of opening of the TolC channel [1] and of substrate extrusion by AcrB [2] separately, we have continued the analysis of the latter one. Thereby, we have focused both on the local interactions between substrate and protein, the properties of the extrusion pathway, as well as the principal subdomain movements which lead to the peristaltic motion. Furthermore, we have investigated the possibility to pull the substrate from the final state of the previous simulations out of the exit gate to estimate whether the substrate is already free to leave the protein via diffusion, which is usually beyond the time scale of computer simulations.

[1] R. Schulz, U. Kleinekathöfer, Biophys. J. 96, 3116 (2009)

[2] R. Schulz, A.V. Vargiu, F. Collu, U. Kleinekathöfer, P. Ruggerone, submitted

BP 7.9 Mon 17:15 Poster B1

Synchronisation in a Chain of Rowers with Hydrodynamic Interaction — ●CHRISTOPHER WOLLIN and HOLGER STARK — TU-Berlin, Sekr. EW 7-1, Inst. f. Theo. Physik, Hardenbergstr. 36, D-10623 BERLIN-Charlottenburg

The ciliary beat, for example of paramecium and opalina, is coordinated such that metachronal waves move along the cell surface. There is strong evidence that hydrodynamic interactions cause these waves.

In order to study the origin of metachronal waves, we investigate the collective dynamics of a chain of periodically moving beads, called rowers, which are to abstract the ciliary beat. The beads move on line segments situated close to an infinitely extended planar wall. They are driven by a force that possesses a quadratic potential and that is reversed when the bead reaches a given amplitude in each direction. We assume the beads to be pointlike and describe their hydrodynamic interaction by the Blake tensor. Varying the distance of the segments from the wall, we can tune the range of the hydrodynamic interaction.

We find that two rowers synchronize in phase or in anti-phase depending on the respective negative or positive curvature of the driving quadratic potential. Chains with more rowers display a wealth of self-organized pattern formation. In particular, in the case where two rowers would synchronize in phase, we observe stable metachronal waves when the chain is located close to the wall, i.e., when the hydrodynamic interaction predominantly acts between nearest neighbours. Moving the chain away from the wall, the metachronal waves disappear and only transient structures form.

BP 7.10 Mon 17:15 Poster B1

Dynamic length regulation of microtubules — ●LOUIS REESE, ANNA MELBINGER, and ERWIN FREY — Arnold Sommerfeld Center for Theoretical Physics (ASC) and Center for NanoScience (CeNS), Department of Physics, Ludwig-Maximilians-Universität München

Microtubules are highly dynamic filaments that perform a variety of tasks in living cells. They serve as intracellular highways for molecular motors, which are transported along those tracks or diffuse in the cytosol. Here we examine mechanisms to regulate microtubule-length through the concentration of motors in the cell [1]. It is analyzed how the interplay between density-dependent transport on the tracks, and filament polymerization affects the dynamics of filament length. Employing stochastic simulations complemented by analytic calculus we identify distinct dynamic regimes. The model presented is conform with recent experiments studying in vitro microtubule depolymerization [2]. Our findings show that molecular motors can specifically control MT length fluctuations.

[1] A. Parmeggiani, T. Franosch, E. Frey, Phys. Rev. Lett. 90, 086601 (2003).

[2] V. Varga, J. Helenius, K. Tanaka, A.A. Hyman, T.U. Tanaka and J. Howard, Nat. Cell Biol. 8, 957 (2006)

BP 7.11 Mon 17:15 Poster B1

Study of H/D substitution effects on the function of the cytochrome bc_1 complex of *Rhodobacter capsulatus* — ●KATRIN JAHNS¹, NATALIA VOSKOBOYNIKOVA¹, MARIA KOZLOVA^{1,2}, and ARMEN MULKIDJANIAN^{1,2} — ¹School of Physics, University of Osnabrück, D-49069 Osnabrück, Germany — ²A.N.Belozersky Institute of Physico-Chemical Biology, Moscow State University, Moscow, 119991, Russia

The cytochrome bc_1 complex is a voltage-generating membrane ubiquinol:cytochrome c oxidoreductase [1]. We have studied the effect of the $\text{H}_2\text{O}/\text{D}_2\text{O}$ substitution on the flash-induced turnovers of the cytochrome bc_1 complexes in the vesicular preparations of the inner cellular membranes (chromatophores) of phototrophic α -proteobacteria *Rhodobacter capsulatus*. We traced the kinetics of flash-induced generation of membrane voltage by the cytochrome bc_1 complex via the spectral shifts of native carotenoid pigments and correlated them with the kinetics of electron transfer as measured in the same samples. At neutral pH, the kH/kD ratio was ca. 2.3, it dropped below 2 at acidic and alkaline pH. On contrast, the rates of flash-induced cytochrome b reduction were only ca. 1.5 times slower in D_2O than in H_2O . We conclude that, at physiological pH values, the rate of proton translocation in the cytochrome bc_1 complex is limited by the breakage or formation of hydrogen bonds and not by the transmembrane electron transfer in cytochrome b .

[1] A.Y. Mulkidjanian, Photochem Photobiol Sci 6 (2007) 19-34