BP 12: Cellular Force Generation

Time: Tuesday 17:30–18:45 Location: C 243

BP 12.1 Tue 17:30 C 243

Force and Motorprotein Concentration Determine Dynamics of Bacterial Pili — •Martin Clausen and Berenike Maier — WWU Münster, Institut für Allgemeine Zoologie und Genetik, Schloßplatz 5, 48149 Münster, Germany

Type IV pili are major bacterial virulence factors required for adhesion, surface motility and gene transfer. In the human pathogen Neisseria gonorrhoeae, these flexible polymeric filaments extend several micrometers from the cell surface and generate force in the range of 100pN by retraction. Two antagonistic ATPases, PilF and PilT, support elongation and retraction respectively. We investigated the dynamics of individual pili using laser tweezers and observed that the probability for polymerization increased with increasing force for forces up to 100pN. The length change of the pilus as a function of time was analyzed using the statistical randomness parameter as well as direct sectioning. The data reveals two distinct time scales: on a time scale of miliseconds backsteps and pauses were detected, while on the longer timescale directional reversal of the pilus movement was observed. This observation is inconsistent with simple Arrhenius kinetics. We therefore investigated the effect of the concentration of the pilus retraction ATPase PilT and found that the retraction probability decreased with decreasing PilT concentration indicating that binding of PilT strongly increases the retraction probability. Fine-tuning of pilus dynamics by force and motor concentration may be important for surface motility and interaction with mammalian cells.

BP 12.2 Tue 17:45 C 243

Efficiency of molecular motors at maximum power: "Power stroke" beats "Brownian ratchet" — •Tim Schmiedl and Udo Seifert — II. Institut für Theoretische Physik, Universität Stuttgart, 70550 Stuttgart

Molecular motors transduce chemical energy from hydrolizing ATP into mechanical work exerted against an external force. The efficiency of such a motor usually increases when increasing the force, reaching the maximum at the stall force. At this force, however, the velocity of the molecular motor and the power output vanishes. It is thus more meaningful to characterize such motors by the efficiency at maximum power. We calculate this efficiency for a simple model and show that the qualitative behaviour depends crucially on the position of the transition state or, equivalently, on whether the motor step occurs in a so called "power stroke" or in a "Brownian ratchet" manner. Specifically, we find a power stroke mechanism, as realized e.g. in myosin motors, to be most favourable with respect to both high power output and high efficiency at maximum power. In this regime, driving the motor farther out of equilibrium by applying higher chemical potential differences can, contra-intuitively, increase the efficiency.

BP 12.3 Tue 18:00 C 243

Transport of micrometer-sized vesicles by kinesin in vitro — •Christoph Herold[1], Cécile Leduc[2], Eugene P. Petrov[1], Stefan Diez[2], and Petra Schwille[1] — [1]Biophysics / BIOTEC, TU Dresden, Tatzberg 47-51, 01307 Dresden — [2]Max Planck Institute of Molecular Cell Biology and Genetics, Pfotenhauerstr.

Cytoskeletal motor proteins (e.g., kinesin) are responsible for directed transport in cells. Motor proteins can also be used in artificial bionanotechnological systems to provide a controlled cargo transport. We explore this possibility by using giant unilamellar vesicles (GUVs) as a micrometer-sized cargo model and establish an *in vitro* system to transport this cargo by kinesin (rK430) molecules along surface-attached microtubules (MTs). Kinesin was linked to GUVs (diameter 1−4 μm) via biotin−streptavidin interaction. MTs and moving GUVs were visualized using fluorescence wide-field imaging microscopy. We observe directed transport of GUVs along MTs with traveling distances of up to 100 μm and velocities of $\sim$0.7 μm/s being in a good agreement with the velocity of kinesin motion along MTs ($\sim$0.8 μm/s). The long walking distances, as well as the visualization of the GFP-labeled kinesin molecules by total internal reflection fluorescence imaging, suggest that a large number ($\gtrsim$10) of kinesin molecules is involved in the transport of a single GUV. Apart from its biotechnological importance, this system might additionally be useful to gain further understanding of vesicle transport processes in cells.

BP 12.4 Tue 18:15 C 243

Stochastic force generation by small ensembles of myosin II molecular motors — Thorsten Erdmann[1] and •Ulrich Schwarz[2] — [1]Amolf, Biochemical Networks Group, 1098 SJ Amsterdam, The Netherlands — [2]University of Heidelberg, Bioquant 0013, INF 267, 69120 Heidelberg, Germany

Myosin II molecular motors are non-processive and therefore have to work together in ensembles in order to generate appreciable levels of force. In contrast to the situation in muscle, in the actin cytoskeleton (including the actin cortex and stress fibers) myosin II molecular motors usually work in small groups and therefore stochastic effects are expected to be more pronounced. Taking advantage of the separation of time scales present in the myosin II hydrolysis cycle, we are able to reduce the complex network of stochastic transitions within a finite-sized ensemble of myosin II motors to a one-step master equation. We derive analytical expressions for the average time of attachement and the average walk length. We also derive force-velocity relations as a function of ensemble size and compare the average results with exact stochastic simulations of single realizations. Our results show that stochastic effects persist up to a system size of about 15 motors.

BP 12.5 Tue 18:30 C 243

Interaction with host cells influences bacterial pilus dynamics — •Dirk Opitz and Berenike Maier — WWU Münster, Institut für Allgemeine Zoologie und Genetik, Schloßplatz 5, 48149 Münster, Germany

The human pathogen *Neisseria gonorrhoeae* generates force in the range of 100pN. The force generating machine is the type IV pilus, a polymeric cell appendage that generates force by retracting into the cell body. Eucaryotic cells can sense mechanical force and respond by cytoskeletal rearrangements. We therefore hypothesize that force generated by pilus retraction is a signal to their epithelial host cell which may facilitate phagocytosis. Vice versa, products of activated signalling pathways upregulate the expression level of the putative pilus retraction motor PilT. Here, we investigated the dynamics and force generation by individual type IV pili using laser tweezers between 3h and 24h after infection of epithelial cells was initiated. We found that the velocity at forces below 50pN decreased from (1.1±0.1)μm/s in abiotic environment to (0.7±0.1)μm/s. Bacteria generated considerable force during infection but the maximum force was reduced from (125±36)pN in abiotic environment to (73±25)pN on epithelial cells independent of infection time. The type IV pilus dynamics in abiotic environment and on host cells is significantly different suggesting that the signalling between pathogen and host cell is bidirectional.