BP 36: Cytoskeletal Filaments I

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

BP 36.1 Thu 15:00 SCH A251

Lattice defects induce microtubule self-renewal — Laura Schaedel[1], Sarah Triclin[1], Denis Chrétien[2], Ariane Abrieu[3], Charlotte Aumeier[1], Jérémie Gaillard[1], Laurent Blanchoin[1,4], Manuel Théry[1,4], and •Karin John[5] — [1]Univ. Grenoble-Alpes, CEA, CNRS, INRA, Biosciences & Biotechnology Institute of Grenoble, Laboratoire de Physiologie Cellulaire & Végétale, CytoMorpho Lab, 38054 Grenoble, France — [2]Univ. Rennes, CNRS, IGDR (Institute of Genetics and Development of Rennes) - UMR 6290, F-35000 Rennes, France — [3]CRBM, CNRS, University of Montpellier, Montpellier, France — [4]Univ. Paris Diderot, INSERM, CEA, Hôpital Saint Louis, Institut Universitaire d'Hematologie, UMRS1160, CytoMorpho Lab, 75010 Paris, France — [5]Univ. Grenoble-Alpes, CNRS, Laboratoire Interdisciplinaire de Physique, 38000 Grenoble, France

Microtubules are dynamic polymers, which grow and shrink at their extremities. Within the microtubule shaft, tubulin dimers adopt a highly ordered lattice structure, which is generally not considered to be dynamic. Here we report a new aspect of microtubule dynamics, whereby thermal forces are sufficient to remodel the lattice, despite its apparent stability. Our combined experimental data and numerical simulations on lattice dynamics and structure demonstrate that dimers can spontaneously leave and be incorporated into the lattice at structural defects. We propose a model mechanism, where the lattice dynamics is initiated via a passive breathing mechanism at dislocations, which are frequent in rapidly growing microtubules.

BP 36.2 Thu 15:15 SCH A251

Hidden Dynamics of the Red Blood Cell Cytoskeleton — •Julia Jäger[1,2], Michael Lanzer[3], and Ulrich S Schwarz[1,2] — [1]Institut für Theoretische Physik, Universität Heidelberg — [2]Bioquant, Universität Heidelberg — [3]Parasitologie, Universitätsklinikum Heidelberg

The spectrin-actin cytoskeleton of the red blood cell (RBC) is usually considered to be relatively static. Recent studies on malaria infections however have started to change this picture. Malaria parasites invade red blood cells in order to hide from the immune system and to digest hemoglobin. During the time course of the 48 hours until exit, they completely remodel the host cell envelope. This includes dramatic changes of the cytoskeleton, which most likely exploit dynamical processes that have gone unnoticed before. To better understand the dynamics of the cytoskeleton of healthy RBCs, we perform stochastic particle-based computer simulations, which in particular include the polymerization and depolarization of the junctional actin filaments. We then examine different potential mechanisms with which the parasite could exploit these dynamics of the RBC-cytoskeleton to remodel the host cell.

BP 36.3 Thu 15:30 SCH A251

What it takes to become a MAP — •Hauke Drechsler[1], Yong Xu[1], Veikko F. Geyer[1], Yixin Zhang[1], and Stefan Diez[1,2] — [1]B CUBE - Center for Molecular Bioengineering, Technische Universität Dresden, 01307 Dresden, Germany — [2]Cluster of Excellence Physics of Life, Technische Universität Dresden, 01062 Dresden, Germany

The microtubule-binding domains of microtubule-associated proteins (MAPs) are structurally divergent, but often depend on electrostatic interactions with the negatively charged microtubule surface - suggesting that a MAP may primarily be defined by the surface exposure of positive charges rather than by a certain structural fold. Consistently, positively charged artificial objects are able to bind to microtubules and to diffuse along their lattice. Natural MAPs, however, exhibit a more sophisticated functionality beyond lattice-diffusion. Hence, we asked whether basic electrostatic interactions also support advanced MAP functionality. To test this, we studied simple positively charged peptides for the occurrence of typical MAP-like behavior. We found that a multivalent peptide construct featuring four lysine-alanine heptarepeats (starPEG-(KA7)4) shows advanced, biologically relevant MAP-like behavior: starPEG-(KA7)4 binds microtubules in the low nanomolar range, diffuses along their lattice, and tracks depolymerizing microtubule ends. Further, it promotes microtubule nucleation and growth, mediates depolymerization coupled pulling at plus ends, and bundles microtubules without significantly interfering with other proteins on the microtubule. Our results show that positive charges and multivalency are sufficient to mimic advanced MAP-like behavior.

Invited Talk BP 36.4 Thu 15:45 SCH A251

Mechanical properties of intermediate filaments at high strains — Johanna Forsting, Julia Kraxner, Charlotta Lorenz, Anna Schepers, and •Sarah Köster — Institute for X-Ray Physics, University of Göttingen

Mechanical properties of eukaryotic cells are to a great part determined by the cytoskeleton, a composite biopolymer network composed of three filament systems – intermediate filaments, F-actin and microtubules – along with cross-linkers and molecular motors. While actin and tubulin are conserved between cell types and organisms, intermediate filament proteins are expressed in a cell type dependent manner. It has been shown previously that the presence of filaments in a cell has an influence on cell mechanics. Here we unravel the role of the mechanical properties of the individual filaments, in particular at high strains. The molecular architecture of intermediate filaments displays several particularities, such as a strictly hierarchical build-up and multipe alpha-helical domains arranged in parallel. This architecture gives rise to intriguing mechanical properties, such as high flexibility and extreme extensibility. We employ optical traps to obtain precise force-strain data of vimentin and keratin intermediate filaments and model our data by Monte Carlo simulations. We are thus able to show differences between different types of intermediate filaments, as well as a dependence on the ionic anvironment and pH, thus revealing a strong influence of charge interactions.

15 min. coffee break

BP 36.5 Thu 16:30 SCH A251

Direct measurements of interactions between intermediate filaments — •Anna V. Schepers[1], Charlotta Lorenz[1], Stefan Klumpp[2], and Sarah Köster[1] — [1]Insitute for X-Ray Physics, Georg August University Göttingen — [2]Institute for Dynamics of Complex Systems, Georg August University Göttingen

The cytoskeleton consists of F-actin, microtublues and intermediate filaments (IFs), which form a complex composite network. F-actin and microtubule networks have been studied extensively and a large variety of cross-linkers are known. By contrast, the interactions in reconstituted IF networks are less well understood. It has, however, been shown that multivalent ions cause bundling and network stiffening. Whereas rheological experiments give insight into the network properties, it is challenging to distinguish the contributions of filament stiffening and of increased attraction. Combining optical trapping and fluorescence microscopy enables us to bring two single vimentin IFs in contact and directly study the interactions between the filaments. By amplifying electrostatic attraction or diminishing the hydrophobic interactions we are able to study the nature of the interactions between IFs. These results, in combination with studies of the mechanical properties of single IFs, allow us to model the interactions with Monte-Carlo simulations, thereby gaining a deeper understanding of cytoskeletal structures.

BP 36.6 Thu 16:45 SCH A251

Influence of Phosphorylation on Vimentin Mechanics — •Julia Kraxner[1], Julia Menzel[2], Henning Urlaub[3], Blanche Schwappach[2], and Sarah Köster[1] — [1]Institute for X-Ray Physics, University of Göttingen — [2]Department of Molecular Biology, University Medical Center Göttingen — [3]Bioanalytical Mass Spectrometry, Max Planck Institute for Biophysical Chemistry, Göttingen

The mechanical properties of biological cells are determined by the cytoskeleton. This composite biopolymer network consists of microtubules and microfilaments, which are conserved throughout all cell types, and different types of intermediate filaments (IFs), which are expressed in a cell-type specific manner. The adaption to specific mechanical requirements may be further achieved by post-translational modifications of the proteins. In this context, phosphorylation which adds negative charges to the modified site, plays an important role. Regarding IFs, phosphorylation heavily affects disassembly of the filaments and provides binding sites for proteins like 14-3-3 which is a regulator for signaling proteins. Here, we study partially phosphorylated single vimentin IFs by analyzing stress-strain curves recorded with an

optical tweezer setup which combines microfluidics and fluorescence microscopy. Furthermore, we investigate the influence of bound 14-3-3 on the mechanics and the contribution of single phosphorylation sites by phosphomimetics. Our results show that additional charges within the filament soften the vimentin filaments and the binding of 14-3-3 weakens the filaments even more.

BP 36.7 Thu 17:00 SCH A251

Stiffening of the Ndc80 complex, the main mirotubule-kinetochore linker — •Felix Schwietert and Jan Kierfeld — TU Dortmund University, 44221 Dortmund, Germany

In the mitotic spindle microtubules attach to chromosomes via kinetochores, whose molecular structure and mechanical properties are not completely understood. Over the past years, it became evident that the Ndc80 complex plays a major role for attaching microtubules to the kinetochore and transmitting forces from depolymerizing microtubules to the chromosome. The Ndc80 complex is a rod-like coiled-coil with globular end domains that bind to the kinetochore and the microtubule, respectively. Due to its force transmitting function, its elastic properties are of great interest for modeling and understanding chromosome dynamics in the mitotic spindle. Here, we theoretically explain the recent experimental result that the effective stiffness of a Ndc80 complex increases under tension [1]. Our model is based on the specific architecture of the Ndc80 complex, which has a characteristic flexible kink at approximately one third of its length.

[1] V. A. Volkov, P. J. Huis in 't Veld, M. Dogterom, and A. Musacchio, eLife 7:e36764 (2018)

BP 36.8 Thu 17:15 SCH A251

Multiplication of gliding microtubules for biocomputational applications — •Cordula Reuther[1], Paula Santos Otte[1], Rahul Grover[1], Till Korten[1], Günther Woehlke[3], and Stefan Diez[1,2] — [1]B CUBE, TU Dresden, Dresden, Germany — [2]Cluster of Excellence Physics of Life, TU Dresden, 01062 Dresden, Germany — [3]Department of Physics, TU München, Garching, Germany

Recently, an approach to solve combinatorial problems was demonstrated by kinesin-1 driven microtubules exploring, as autonomous agents, physical networks of nanometer-sized channels [Nicolau et al., PNAS, 113(10), 2016]. The possibility to multiply the agents exponentially while traversing such networks is crucial for the scalability of these systems. We developed a method for the multiplication of microtubules gliding on surface-immobilized kinesin-1 and kinesin-14 molecules, respectively. Specifically, our method comprises two simultaneously proceeding processes: (1) elongation of microtubules by self-assembly of tubulin dimers and (2) cutting of microtubules by the severing enzyme spastin. The main challenge in doing so is to optimize both processes such that the average length of the filaments stays roughly constant over time while the number of filaments increases exponentially. Additionally, nucleation of new filaments ought to be avoided in order to prevent errors in the calculations performed by the microtubules. Thus, we first studied each of the two processes separately under various conditions before combining the optimized protocols to actually multiply microtubules. Finally, we aim to multiply microtubules in a physical network with channel structures.