BP 6: Cytoskeletal Filaments I

Time: Monday 15:00–17:30 Location: H 1058

BP 6.1 Mon 15:00 H 1058

Glassy Dynamics in Composite Biopolymer Networks — •Tom Golde[1], Constantin Huster[1], Martin Glaser[1,2], Tina Händler[1,2], Harald Herrmann[3,4], Josef A. Käs[1], and Jörg Schnauss[1,2] — [1]University of Leipzig, Leipzig, Germany — [2]Fraunhofer IZI, Leipzig, Germany — [3]German Cancer Research Center, Heidelberg, Germany — [4]University Hospital Erlangen, Erlangen, Germany

The cytoskeleton is a highly interconnected meshwork of strongly coupled filament systems providing mechanical stability as well as dynamic functions to cells. To elucidate the underlying biophysical principles it is central to investigate not only one distinct functional subsystem but rather their interplay as composite biopolymer structures. Here, we show that composite networks of actin and vimentin filaments can be fully described by a superposition of two non-interacting scaffolds. We demonstrate arising effects in a scale-spanning frame connecting single filament dynamics to macro-rheological network properties and show that linear and non-linear bulk mechanics of actin and vimentin filament networks are captured within the glassy wormlike chain model. Our findings clearly disagree with previous studies reporting emergent effects in these composite networks. These new insights pave the way to deterministically predict the mechanics of the cytoskeleton in distinct cell types based on the properties of its single structural components.[1]

[1] Golde et al., Submitted

BP 6.2 Mon 15:15 H 1058

The mitotic spindle is chiral due to torques generated by motor proteins — Maja Novak[1], •Bruno Polak[2], Juraj Simunic[2], Zvonimir Boban[1], Andreas W. Thomae[3], Iva M. Tolic[2], and Nenad Pavin[1] — [1]Faculty of Science, University of Zagreb, Zagreb, Croatia — [2]Rudjer Boskovic Institute, Zagreb, Croatia — [3]University of Munich, Munich, Germany

Mitosis relies on forces generated in the spindle, a micro-machine composed of microtubules and associated proteins. Forces are required for the congression of chromosomes to the metaphase plate and their separation in anaphase. However, torques may also exist in the spindle, yet they have not been investigated. Here we show that the spindle is chiral. Chirality is evident from the finding that microtubule bundles follow a left-handed helical path, which cannot be explained by forces but rather by torques acting in the bundles. STED super-resolution and confocal microscopy of human spindles revealed that the average helicity of the bundles with respect to the spindle axis is about -2°/μm. Inactivation of kinesin-5 (Kif11/Eg5) abolished the chirality of the spindle. We introduce a theoretical model, which predicts that torques generate curved shapes of bundles, where the twisting component of the torque is required for the helical component of the shape. By comparing the model with experiments, we find that the twisting moment is roughly -10 pNμm. We conclude that torques generated by motor proteins, in addition to forces, exist in the spindle and determine its architecture.

Reference: bioRxiv 167437, https://doi.org/10.1101/167437

BP 6.3 Mon 15:30 H 1058

Metaphase kinetochore movements are regulated by kinesin-8 motors and microtubule dynamic instability — •Agneza Bosilj[1], Anna Klemm[2], Iva Tolic[2,3], and Nenad Pavin[1] — [1]Department of Physics, Faculty of Science, University of Zagreb, Bijenicka cesta 32, 10000 Zagreb, Croatia — [2]Max Planck Institute of Molecular Cell Biology and Genetics, Pfotenhauerstr. 108, 01307 Dresden, Germany — [3]Division of Molecular Biology, Ruder Boskovic Institute, Bijenicka cesta 54, 10000 Zagreb, Croatia

During metaphase, sister chromatids are connected to microtubules (MTs) extending from the opposite spindle poles via kinetochores, protein complexes on the chromosome. Kinetochores congress to the equatorial plane of the spindle and oscillate around it, with kinesin-8 motors restricting these movements. Yet, the physical mechanism underlying kinetochore movements is unclear. We show that kinetochore movements in the fission yeast Schizosaccharomyces pombe are regulated by kinesin-8-promoted MT catastrophe, force-induced rescue and MT dynamic instability. A candidate screen showed that only kinesin-8 motors Klp5/Klp6 are required for kinetochore centering. Our theoretical model with Langevin description of MT dynamic instability shows that kinesin-8 motors are required for kinetochore centering, whereas sensitivity of rescue to force is necessary for the generation of oscillations. We found that irregular kinetochore movements occur for a broader range of parameters than regular oscillations. Thus, our work shows how regulation of MT dynamic instability contributes to kinetochore congression and the accompanying oscillations.

Invited Talk BP 6.4 Mon 15:45 H 1058

Broken detailed balance in active biopolymer assemblies — •Chase Broedersz — Ludwig-Maximilians-Universitaet München, Munich, Germany.

We present a non-invasive approach to identify and quantify non-equilibrium dynamics in living systems based on broken detailed balance. With this approach, we study the dynamics of beating flagella, primary cilia, and cytoskeletal networks. In particular, we use stochastic time traces of the system's dynamics to infer the probability currents in a phase space of the mesoscopic configurational coordinates of a biological assembly. In addition, we will present a more general theoretical framework to investigate what information about the system's non-equilibrium state can be extracted from such phase space currents. For example, we will discuss how to extract the entropy production rate - a measure of the dissipated power in a driven system - from measured current cycles. Next, we present predictions for the scaling behavior of the entropy production rate with the distance between measurement points in the system. Our results provide insight in to how internal driving by enzymatic activity generates non-equilibrium dynamics on different scales in a variety of biological systems, including biopolymers and their assemblies.

BP 6.5 Mon 16:15 H 1058

Stress-Strain Behavior of Vimentin Intermediate Filaments — •Johanna Block[1], Hannes Witt[2], Andreas Janshoff[2], and Sarah Köster[1] — [1]Institute for X-Ray Physics, University of Goettingen, 37077 Göttingen, Germany — [2]Institute of Physical Chemistry, University of Goettingen, 37077 Göttingen, Germany

It is widely accepted that the cytoskeleton, which is composed of three filamentous protein structures - microfilaments (MFs), microtubules (MTs) and intermediate filaments (IFs) - plays a major role for cell mechanics. Whereas MFs and MTs are conserved between cell types, at least 70 different genes in humans code for IFs, which are expressed in a cell type specific manner. So far, it was not possible to infer the mechanical properties found on length scales of protein superstructure, cells and beyond, from the peculiar molecular architecture of IFs. Using optical tweezers, combined with microfluidics and fluorescence microscopy, we directly probed the stress-strain behavior of single vimentin IFs under physiological buffer conditions in a highly controlled fashion. We found a strong loading-rate dependent behavior, indicating that vimentin IFs act as a "safety belt" for cells. Further, our results provide evidence that single vimentin IFs act as an intracellular shock absorber using a balance of classical energy dissipation and storage of potential energy. By theoretical modelling and Monte Carlo simulations we are able to directly attribute filament mechanics to a molecular mechanism and reveal an intriguing non-equilibrium phenomenon leading to pronounced energy dissipation and mechanical adaption.

BP 6.6 Mon 16:30 H 1058

Actin dynamics deform membrane in and out mimicking filopodia and endocytosis — •Camille Simon[1], Remy Kusters[1], Valentina Caorsi[1], Jean-François Joanny[2], Clément Campillo[3], Julie Plastino[1], Pierre Sens[1], and Cécile Sykes[1] — [1]Institut Curie, Paris, France — [2]ESPCI, Paris, France — [3]Université Evry Val d'Essonne, Evry, France

The cell membrane is able to deform inward, as in endocytosis intiation, or outward, as in filopodia formation. Interestingly, both deformations are generated by the same branched, Arp2/3-based, polymerizing actin network. How an inward or an outward deformation can result from the same network structure? What are the physical parameters that will trigger the direction of membrane deformation? To address these questions, we use a reconstituted membrane system of liposomes and purified actin. A dynamic branched actin network is generated at the liposome surface. We investigate the conditions

under which the actin cytoskeleton induces inward or outward membrane deformations. We reveal that actin dynamics is the sole player of membrane deformations by photo-damaging the actin structure that relaxes membrane shape. Lowering membrane tension is key to produce filopodia-like structures. Oppositely, endocytic-like structures are robust features that only weakly depend on membrane tension. A pulse-chase two color actin experiment reveals the details of network growth during inward or outward membrane deformation. Our results, supported by theoretical models, explain how such deformations depend on a mechanical balance between the membrane and the actin network.

BP 6.7 Mon 16:45 H 1058

Dynactin stabilises microtubules to establish their uniform orientation in neuronal axons — •Maximilian Jakobs and Kristian Franze — Department of Physiology, Development and Neuroscience, University of Cambridge, Cambridge, UK

The microtubule (MT) cytoskeleton in neuronal axons is highly oriented with almost all MTs pointing with their growing end (+end) away from the cell body (+end out). Motor proteins rely on this orientation to move cellular cargo to the distal regions of the axon. Despite 30 years of research, the mechanism that establishes MT orientation remains unknown. We here analysed MT growth with supervised machine learning in *D. melanogaster* neurons, complemented by an analytical model of MT growth. We found that +end out MTs grow for longer times than oppositely oriented MTs (−end out). According to our model, this leads to dramatic differences in average MT lengths, so that −end out MTs are short and unstable. Additionally, we found evidence that dynactin is responsible for the differences in growth times by promoting growth at the axonal tip through a molecular gradient. These findings suggest a simple mechanism that organises axonal MTs. First, +end out MTs are stabilized by distally located dynactin. Subsequently, the short −end out MTs depolymerize or reorient, leaving only +end out MTs in the axon. Our results pave the way towards a deeper understanding of how the cytoskeleton in neurons orients to support molecular transport, potentially shedding light on pathologies that are characterized by axonal transport deficiencies such as Alzheimers disease.

BP 6.8 Mon 17:00 H 1058

A field-theoretic approach to microtubule growth — •Johannes Pausch and Gunnar Pruessner — Department of Mathematics, Imperial College London, United Kingdom

Microtubule filaments are a major part of the cytoskeleton. They influence the shape and movement of the cell and are used for transport processes inside the cell. Microtubules grow and shrink by polymerising and depolymerising, that is by absorbing and emitting tubulin which diffusively spread in the cytoplasm. Here, we model the stochastic process of microtubule growth as a field theory.

In our model, we recover the classic diffusion and diffusion-convection results. Furthermore, we are able to model the tubulin-absorption-induced spatially discrete growth of the microtubule filament and find analytic real space results for expected assembly speed and variance. Our approach produces analytic expressions in Fourier space that require a short-length scale cutoff in two dimensions and above. It is particularly flexible to incorporate more complex interactions between microtubules and tubulin. In one dimension, our results are easily compared to corresponding results using probabilistic techniques.

BP 6.9 Mon 17:15 H 1058

Investigations on the cell morphology of oral mucosa cancer and non-cancer cells — •Nina Bartels[1], Maja Strugacevac[1], Constanze Wiek[2], Julia Kristin[2], Marcel Glaas[2], Jörg Schipper[2], and Mathias Getzlaff[1] — [1]Heinrich-Heine-Universität Düsseldorf, Institute of Applied Physics, Universitätsstr. 1, 40225 Düsseldorf, Germany — [2]Düsseldorf University Hospital, Department of Othorinolaringology, Moorenstrasse 5, 40225 Düsseldorf, Germany

In order to develop new cell-selective treatment strategies for head and neck squamous cell carcinoma, our group is investigating the differences of the cell morphology and physical properties of different oral cancer cells and oral keratinocytes. The cell lines originate from different locations of the oral mucosa and are investigated using a fluorescence microscope.

To obtain more information about cell morphology, the cells were stained using CellMask Green (cell membrane) and Hoechst (cell nuclei) staining kits. By the confocal laser-scanning microscope three-dimensional images of the cells were made to compare the different cell lines in size, volume and shape.

A staining kit for active mitochondria (MitoTracker) enables us to compare the aerobic metabolism of tumor and non-cancer cells to verify the Warburg hypothesis. Additionally, actin filaments and microtubules were stained to observe differences in the cytoskeleton which is specific for cell elasticity. This contribution will show and discuss our latest results.