BP 27: Posters: Cytoskeletal Filaments

Time: Thursday 17:30–19:30 Location: Poster A

BP 27.1 Thu 17:30 Poster A

Cell plasticity is tightly linked to elastic stresses in the cytoskeleton — •Achim Schilling, Navid Bonakdar, Michael Kuhn, Richard Gerum, and Ben Fabry — Biophysics Group, FAU

Cells show pronounced non-linear visco-elastic and visco-plastic properties under large deformations and forces that are important for protecting the cell against mechanical damage. We used a high-force magnetic tweezer setup to deliver unidirectional forces with high precision of up to 30 nN to fibronectin-coated magnetic 5um beads bound to cell surface adhesion receptors. To probe cells with bidirectional forces, the cell culture plate was placed on a rotational/translational stage such that the magnetic bead remained at a constant distance to the magnetic tweezer tip after a 180° rotation. Bead displacements were measured during application of force steps (creep response) and after the force was removed (recovery response). With increasing force magnitude, the cells stiffened, and the recovery became increasingly incomplete, indicating the emergence of plastic behavior. This plasticity was a constant fraction (20%) of the total bead displacement. We attribute the plastic behavior to a buildup of excess slack in the cytoskeletal fibers; when the force direction was suddenly reversed, the beads jumped by twice the slack length in the opposite direction. The creep and the recovery response were fully characterized by a simple power-law vs. time with only 2 force-dependent parameters (elasticity and creep exponent). Our results show that plastic energy dissipation during large cell deformations is tightly linked to elastic stress dissipation and provides additional protection against mechanical damage.

BP 27.2 Thu 17:30 Poster A

Molecular motors can sharpen the length distribution of treadmilling filaments — •Denis Johann, Christoph Erlenkämper, and Karsten Kruse — Theoretische Physik, Universität des Saarlandes, Postfach 151150, 66041 Saarbrücken, Germany

The assembly of actin filaments and microtubules depends on the hydrolysis of nucleotide tri-phostohates. Together with their structural polarity this can lead to treadmilling, a process during which the filaments, on average, grow at one end and shrink at the other. The distribution of proteins binding to a treadmilling filament increases towards the shrinking end. For proteins affecting the removal rate of filament subunits such a gradient implies an effectively length-dependent depolymerization rate, which can lead to a unimodal length distribution unknown to polymers at equilibrium [1]. Using Monte-Carlo simulations, we show that the width of the length distribution can narrow substantially if the depolymerizing proteins are molecular motors, moving directionally towards the shrinking end. We present expressions for the width of the length distribution in the limits of vanishing and infinite motor speeds.

[1] C. Erlenkämper, K. Kruse, *Phys. Biol.* **6**, 046016 (2009)

BP 27.3 Thu 17:30 Poster A

Length Regulation is an Intrinsic Property of Treadmilling Actin Filaments — •Christoph Erlenkämper and Karsten Kruse — Theoretische Physik, Universität des Saarlandes, 66041 Saarbrücken

Actin polymers constitute a major part of the cell cortex. The polymerisation of these filaments relies on the consumption of chemical energy through the hydrolysis of Adenosin-tri-phosphate (ATP). Together with the structural polarity of actin monomers, this can lead to treadmilling. In this state, the monomers are removed from one end of the filament at the same rate at which they are added at the other end. To investigate the conditions of treadmilling and the accompanying actin-length distribution, we formulate a three-state model of actin dynamics. It accounts for random dephosphorylation of actin-bound ATP along filaments and for the dependence of the various assembly rates on the nucleotide bound to actin monomers. For a range of experimentally accessible parameters, we find that treadmilling goes along with a stationary unimodal length distribution, which is due to a length-dependent monomer removal rate. In this case, the spontaneous disappearance of filaments is largely suppressed. We present analytical estimations of the typical filament length as well as for the variance of the ensuing length distributions and dicuss possible scenarios for *in vivo* length regulation.

BP 27.4 Thu 17:30 Poster A

Cortical dynamics with viscoelasticity — •Arnab Saha[1], Stephan Grill[1,2], Guillaume Salbreux[1], and Frank Jülicher[1] — [1]Max Planck Institute (Physics for complex system) — [2]Max Planck Institute (Molecular cell biology and Genetics)

The cell cortex is a thin layer beneath the cell membrane that largely consists of cross-linked actin filaments, non-muscle myosin motor-protein and various actin binding proteins (ABPs). It can actively contract and generate stress in presence of ATP-consuming motor proteins and treadmilling processes, that drive the cortex far away from thermal equilibrium. Here we present a two dimensional hydrodynamic model for cell cortex valid at long length scale and short to long time scales (incorporating both elastic and viscous regimes), using basic symmetry principles and conservation laws. Using the model, then we probe the elastic regime of cortical dynamics along with the experimental observations from laser ablation of cortex.

BP 27.5 Thu 17:30 Poster A

Measurements of F-actin tube conformation — Evelin Jaschinski, Inka Lauter, Bernd Hoffmann, and •Rudolf Merkel — Forschungszentrum Jülich GmbH, Wilhelm-Johnen-Straße, 52428 Jülich

Actin is a major component of the cytoskeleton of almost all eukaryotic cells. In thermal equilibrium in vitro polymerized actin filaments (F-actin) are fluctuating. These fluctuations are restricted by surrounding filaments which form a tube like volume. The propability distribution of the tube dimension has been measured by means of confocal fluorescence microscopy in our group [1, 2]. Based on these results we are analyzing the F-actin tube conformation under specific external conditions like enforced alignment exerted by shear force as well as counterions.

[1] J.Glaser, D. Chakraborty, K. Kroy & I. Lauter, M. Degawa, N. Kirchgeßner, B. Hoffmann, R. Merkel, M. Giesen, Phys. Rev. Lett. 105, 037801 (2010)

[2] M. Romanowska, H. Hinsch, N. Kirchgeßner, M.Giesen, M. Degawa, B. Hoffmann, E. Frey, R. Merkel, EPL 86, 26003 (2009)

BP 27.6 Thu 17:30 Poster A

Dynamics of semiflexible ring polymers in shear fow — •Philipp Lang and Erwin Frey — Arnold Sommerfeld Center for Theoretical Physics, Ludwig-Maximilians-Universität München

The dynamics of semiflexible ring polymers is studied both in equilibrium and subject to external shear flow. We give a comprehensive analytical description of the relaxational dynamics of internal and global modes in the case of persistence lengths greater or equal than the contour lengths. A relaxation behavior similar to linear polymers is found, yet crossover times and amplitudes are different due to smaller internal fluctuations.

The non-equilibrium dynamics in shear flow is investigated by Brownian dynamics simulations of inextensible rings and interpreted in terms of the relaxational dynamics. We find two distinct types of tumbling events: rapid turnover, and tread-milling in a metastable state. The main tumbling frequency is found to scale with the Weissenberg number $Wi = \kappa \cdot \tau_L$ as $Wi^{3/4}$ with the time scale τ_L given by the global rotational relaxation time.

BP 27.7 Thu 17:30 Poster A

How a polymer breaks a bond — •Sebastian Sturm, Jakob Bullerjahn, and Klaus Kroy — Institut für theoretische Physik, Universität Leipzig, Vor dem Hospitaltore 1, 04103 Leipzig

The unbinding kinetics of crosslinking proteins is a crucial ingredient to the rheological behavior of reversibly crosslinked polymer networks and cells. Whereas current models of cell rheology do account for the influence of crosslinker bindings on single-filament dynamics [1,2], the converse effect of polymer dynamics on unbinding kinetics has so far received less attention. Here we address this issue by investigating the unbinding kinetics of polymer-bound crosslinkers based on a rigorous description of subdiffusive monomer dynamics in terms of memory friction [3].

[1] L. Wolff, P. Fernandez, and K. Kroy, New Journal of Physics (2010)
[2] O. Lieleg, K. M. Schmoller, M. M. A. E. Claessens, and A. R. Bausch, Biophysical Journal 96, 4725-4732 (2009)

[3] J. Bullerjahn, S. Sturm, L. Wolff, and K. Kroy, epl (2011), doi 10.1209/0295-5075/96/48005

BP 27.8 Thu 17:30 Poster A

The role of fluctuations in cytoskeletal wave formation — •Florimond Collette, Marc Neef, and Karsten Kruse — Theoretische Physik, Universität des Saarlandes, Postfach 151150, 66041 Saarbrücken, Germany

Spontaneous cytoskeletal waves have been reported in a number of different cell types and under various conditions. Actin waves observed in the slime mold *Dictyostelium discoideum* [1] and in human neutrophils [2] presumably originate from similar mechanisms: Proteins nucleating actin filaments get activated on the membrane, generate new filaments, and are in turn inactivated by actin filaments through an unknow mechanism. We extend a meanfield description of this system [3] to consider stochastic effects in the dynamics of filament nucleators and solve the equation numericaly. When varying the number of nucleators, we observe bifurcations of stationary states into travelling waves. In the case of travelling waves, we find spontaneous switching between waves moving into opposite directions.

[1] T. Bretschneider, *et al.*, *Curr. Biol.* **14**, 1 (2004).
[2] O. D. Weiner, *et al.*, *PLoS Biology* **5**, e221 (2007).
[3] K. Doubrovinski and K. Kruse, *EPL* **83**, 18003 (2008).

BP 27.9 Thu 17:30 Poster A

Active Microrheology: A new approach to determine mechanical properties of assembled networks — •Tobias Paust[1], Ines Martin[1], Michael Beil[2], Harald Herrmann[3], and Othmar Marti[1] — [1]Institute for Experimental Physics, Ulm University, Ulm, Germany — [2]Internal Medicine I, Ulm University, Ulm, Germany — [3]German Cancer Research Center, Heidelberg, Germany

Macro- and microrheology is extensively used to characterize complex networks of biopolymers. From these data one infers mechanical properties affecting migration or the response to external stresses. So far macro- and microrheology give similar but not identical responses. The reason for this is still under debate.

In this work we explore the possibility of two point microrheology to determine locally the complex tensorial elastic response of heterogeneous networks. We use the measurements to find possible differences between macro- and microrheology. A careful analysis of the data provides additionally the frequency response of the complex elastic tensor.

As a test system we have investigated keratin 8/18 network extracted from epithelial pancreatic cancer cells. We compare the data to measurements of in vitro assembled keratin 8/18 networks.

BP 27.10 Thu 17:30 Poster A

Super-resolution study of paracrystalline actin in auditory receptor cells — •Valeria Piazza, Britta Weinhausen, Tim Salditt, and Sarah Köster — Institute for X-Ray Physics & CRC Physics, University of Göttingen, Germany

In most cell types microfilaments coalesce into networks and bundles with variable degrees of orientation and size. In statoacustical receptor cells, however, actin bundles inside stereocilia show an unusually "extreme" level of organization: thousands of filaments are tightly packed in a paracrystal array to form stiff cylinders (200-500 nm in thickness and with lengths ranging from 2 to 30 μm) with a supportive function for mechanotransduction. We study the peculiar packing symmetry and molecular composition of these cell protrusions by means of fluorescence nanoscopy and X-ray diffraction. With STED (STimulated Emission Depletion microscopy) coupled to immunofluorescence we test when and where each of the two actin isoforms - β and γ - is incorporated in cochlear stereocilia throughout development. Moreover, using spatially-resolved X-ray nano-diffraction we characterize the spacing of the actin filaments within individual stereocilia. The X-ray measurements are performed on hair cells that are chemically fixed in different conditions (for example, different developmental stages), to test whether the filament array changes or it is constantly maintained in one specific state. Eventually the imaging data will be combined with the X-ray analysis to correlate molecular composition to structure in discrete regions of stereocilia.

BP 27.11 Thu 17:30 Poster A

Network elasticity of stiff rods with semi flexible cross-linkers: simulation and experiment — •Meenakshi M Prabhune[1], Knut M Heidemann[2], Florian Rehfeldt[1], Max Wardetzky[2], and Christoph F Schmidt[1] — [1]Drittes Physikalisches Institut,Georg-August-Universität Göttingen, Göttingen — [2]Institute of Num. and Appl. Math, Georg-August-Universität Göttingen, Göttingen

Mechanical stress plays crucial roles in cellular functions such as adhesion, cell division, motility and many others. Experimental approaches, such as the use of elastic substrates, exist to estimate the forces exerted at the boundaries of cells. Mapping cell-internal stress fields still remains a challenge owing to the heterogeneity of the cytoskeletal filaments and to a lack of appropriate sensors. To model a strongly heterogeneous composite, we construct networks of microtubules cross-linked by dsDNA of variable length. We measure the linear and non-linear shear-elastic response in these networks by macro- and microrheology. Simultaneously, we compare the experimental data to numerical simulations that we have developed for networks of stiff slender rods connected by semi-flexible linkers.

BP 27.12 Thu 17:30 Poster A

Vimentin Filaments in Small Configuration Spaces — •Bernd Nöding, Susanne Bauch, and Sarah Köster — Institute for X-Ray Physics and CRC Physics, University of Göttingen, Germany

The eukaryotic cytoskeleton mainly consists of three types of fibrous proteins. While microtubules and microfilaments are highly conserved, intermediate filaments (IFs) vary from one cell type to another. Here, we focus our study on vimentin IFs. Investigations of the mechanical properties of individual filaments are a necessary prerequisite for a better understanding of the mechanics of biopolymer networks and eventually whole cells. The mechanical rigidity of a polymer is characterized by its persistence length L_p. We perform time-resolved measurements of fluorescently labeled filaments in solutions of different viscosities without any interaction with a substrate whatsoever. To this end, we confine the filaments in microchannels of variable widths and heights ($\sim \mu$m). The purpose of using the channels is threefold: first, they mimic the crowded environment of the cell. Second, they reduce the configuration space occupied by the filaments, thereby markedly improving statistics. Third, they realize the Odijk confinement regime. We find that IFs behave as ideal worm-like chains. Inclusion of data for microfilaments, which are also confined in the Odijk regime, shows that both filament systems behave according to a single scaling law. Furthermore, we find that fluctuations in perpendicular directions decouple as predicted by the worm-like chain model. For freely fluctuating individual vimentin filaments we determine $L_p \sim 2$ μm.

BP 27.13 Thu 17:30 Poster A

Formation of regular actin bundle networks by counter-ion condensation and entropic forces — Florian Huber, •Dan Strehle, Jörg Schnauss, Matti Gralka, and Josef Käs — Universität Leipzig

Actin is a major constituent that contributes mechanical integrity and function to living cells. Filamentous actin is organized in structures spanning networks of filaments, bundles of filaments to networks of bundles. In this work we explore the mechanisms that determine this resulting architecture. In our experimental bottom-up approach we combine actin filaments with uniform attractive interactions. Counterion-condensation as well as depletion-force agents aggregate a homogeneous F-actin solution into regular actin-bundle networks connected by aster-like centers. Both, partial nematic or flow-induced alignment, drastically change the observed network architecture to ladder-like patterns. Complementing the experimental data we aim at elucidating the mechanism on the basis of coarse-grained modeling. Due to the fundamental nature of the interactions considered, we expect that the investigated type of network formation further implies severe consequences to cytoskeletal network formation on the more complex level of living cells.

BP 27.14 Thu 17:30 Poster A

Contractile force generation by entropic softening of actin networks — •Carsten Schuldt, Tom Golde, and Josef Käs — Universität Leipzig

One major constituent of the cell's cytoskeleton is the globular protein actin, organized in filaments subjected to steady assembly and disassembly. We study the depolymerization of actin bundles of cross-linked single actin filaments and networks. In particular, the forces associated with this process are of interest.

Retraction at the rear of a cell is a fundamental part of its migration process. This contraction can be accomplished by actin-myosin interaction. However, myosin knock-out cells have been shown to be still capable of migrating. Alternatively, the depolymerization of the cytoskeleton was proposed to cause contractile forces only by a gain in entropy in the absence of molecular motors. This concept has been demonstrated on polymer meshworks of nematode's major sperm protein [1].

We employ a microrheology approach in conjunction with severing proteins to measure both softening and contraction of the depolymerizing network.

[1] Wohlgemuth et al., Biophys. J, 88, 2462 (2005)

BP 27.15 Thu 17:30 Poster A

Diameter of Keratin 8/18 — •Ines Martin[1], Anke Leitner[1], Masoud Amirkhani[1], Vladislav Krzyzanek[2], Michael Beil[3], Harald Herrmann[4], Paul Walther[5], and Othmar Marti[1] — [1]Institute for Experimental Physics, Ulm University, Ulm — [2]Institute of Medical Physics and Biophysics, Münster University, Münster — [3]Internal Medicine I, Ulm University, Ulm — [4]Division of Molecular Genetics, German Cancer Research Center, Heidelberg — [5]Central Facility of Electron Microscopy, Ulm University, Ulm

The cytoskeleton of epithelial cells consists of three types of filament systems: microtubules, intermediate filaments (IFs) and actin filaments. In our work, we have a closer look at intermediate filaments, which are responsible for the stiffness of cells and responses to mechanical stimuli.

Because of their heterogeneous nature, it is not exactly known how IFs are arranged over their cross-section, even though some measurements of diameter, mass per length and cross-section exist. Therefore we compared the diameter of filaments with different techniques: Transmission Electron Microscopy with negative staining, Scanning Electron Microscopy of freeze-dried samples with tungsten coating, Scanning Transmission Electron Microscopy (STEM) with and without glutaraldehyde fixation and Small Angle X-ray Scattering (SAXS). Additionally we studied the behavior of Keratin 8/18 adsorbed on a substrate with SEM of freeze-dried samples. Here we could see flattening of some of the filaments, from which one can conclude in agreement with STEM data that the IFs are not very densely packed.

BP 27.16 Thu 17:30 Poster A

Shear-induced wrinkling in random and regular semiflexible polymer networks — •Pascal Müller and Jan Kierfeld — Physics Department, TU Dortmund, Dortmund

Networks of semiflexible polymers such as F-actin are one of the main components in the cytoskeleton and determine the elastic properties of the cell.

We study the elastic properties of filamentous networks by energy minimisation of deformed two-dimensional networks with regular and random geometries. The planar two-dimensional networks are embedded in three-dimensional space, and our main focus is the investigation of wrinkling under shear stress by enabling transversal displacements of the network sheet. We compare triangular networks to random networks in order to identify the impact of network geometry on different wrinkle properties such as amplitude, wavelength, and the critical shear stress that is required to induce wrinkling.