BP 25: Posters: Cytoskeleton

Time: Wednesday 17:30-19:30

BP 25.1 Wed 17:30 Poster C Molecular Motors Can Sharpen the Length Distribution of

Treadmilling Filaments — •CHRISTOPH ERLENKÄMPER^{1,2}, DENIS JOHANN¹, and KARSTEN KRUSE¹ — ¹Universität des Saarlandes, Saarbrücken, Germany — ²Institut Curie, Paris, France

The assembly of actin filaments and microtubules depends on the hydrolysis of nucleotide tri-phosphates. 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. 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.

BP 25.2 Wed 17:30 Poster C $\,$

A phase-field model for amoeboid motility — •ALEXANDER DREHER¹, IGOR ARONSON^{2,3}, and KARSTEN KRUSE¹ — ¹Theoretische Physik, Universität des Saarlandes, Postfach 151150, D-66041 Saarbrücken, Germany — ²Materials Science Division, Argonne National Laboratory, 9700 S. Cass Avenue, Argonne, IL 60439, USA — ³Engineering Sciences and Applied Mathematics, Northwestern University, 21345 Sheridan Road, Evanston, IL 60202, USA

The crawling of eukaryotic cells on substrates is driven by the cytoskeleton. How the cytoskeleton is organized in this process is still poorly understood. It has been suggested that spontaneous polymerization waves provide a possible answer to this question. We examine this possibility theoretically by analyzing a system of treadmilling filaments in presence of nucleating proteins. A challenge arises from the need to describe a moving deformable cell boundary. In this minimal system we treat the cell shape by a phase-field approach. We find spiral waves as well as self-sustained motion of the cell in agreement with experiments on amoeboid motility.

BP 25.3 Wed 17:30 Poster C **A mechanism of stress generation in contractile rings** — •ANNE WALD¹, VIKTORIA WOLLRAB², DANIEL RIVELINE², and KARSTEN KRUSE¹ — ¹Universität des Saarlandes, Theoretische Physik, Postfach 151150, 66041 Saarbrücken — ²Laboratory of Cell Physics, ISIS/IGBMC, Université de Strasbourg and CNRS (UMR 7006), 8 allée Gaspard Monge, 67083 Strasbourg, France and Development and Stem Cells Program, IGBMC, CNRS (UMR 7104), INSERM (U964), Université de Strasbourg, 1 rue Laurent Fries, BP10142, 67400 Illkirch, France

Cytokinesis is the final step of cell division during which the mother cell is split into her daughters. In many cell types, this process is driven by the contraction of a ring composed of actin filaments and myosin motors. How the interaction between motor molecules and filaments generates the stress necessary for contraction is still poorly understood. We study a possible mechanism based on the existence of bipolar fila- ment structures. These structures emerge, for example, when filaments grow into opposite directions from a common nucleator as has been suggested for actin filaments in fission yeast. We study the dynam- ics of dynamic polar filaments in the presence of molecular motors by using a continuum mean-field description. We calculate the stress generated by a homogenous ring in steady state and investigate the stability of this structure against perturbations. Notably, we find that filament assembly can heal defects that might otherwise lead to ring rupture.

BP 25.4 Wed 17:30 Poster C Depolymerization of Microtubules by Diffusing Motor Proteins — •EMANUEL REITHMANN, LOUIS REESE, and ERWIN FREY — Arnold Sommerfeld Center for Theoretical Physics (ASC) and Center for NanoScience (CeNS), Department of Physics, Ludwig-Maximilians-Universität München

Microtubules (MT) are dynamic protein filaments integral to a wide

Location: Poster C

variety of cellular processes such as mitosis, meiosis and cellular transport. Molecular motors moving on the MT play a key role in the control of MT length as they can catalyze depolymerization and polymerization. There is a plethora of molecular motors with different properties such as biased or unbiased movement along the MT. As opposed to depolymerases with a directed motion, only a few theoretical results exist for diffusing motors such as MCAK. Here we investigate a stochastic model for the processive depolymerase activity induced by molecular motors diffusing on the MT. Employing Monte Carlo simulations and analytic mean-field approximation methods we study the depolymerization process. We determine the stationary density profiles of MCAK along microtubules, and show that the concentration of bound motor proteins at the microtubule tip is a critical determinant of the depolymerization rate. Our model agrees well with experimental results [1]. Further, based on this data, predictions of the MCAK off-rate at the MT tip can be made.

 J. Cooper, M. Wagenbach, C. Asbury, L. Wordeman, Nat. Struct. Mol. Biol. 17, 77-82 (2010).

BP 25.5 Wed 17:30 Poster C Dynamics and length distribution of microtubules under force and confinement — BJÖRN ZELINSKI, •NINA MÜLLER, and JAN KIERFELD — Physics Department, TU Dortmund, Dortmund, Germany

We investigate the microtubule polymerization dynamics with catastrophe and rescue events for three different confinement scenarios: (i) The microtubule is confined by rigid and fixed walls, (ii) it grows under constant force, and (iii) it grows against a linearly increasing force. (i) In confinement, we find exponentially decreasing or increasing stationary microtubule length distributions instead of bounded or unbounded phases and introduce a model for wall-induced catastrophes. (ii) Under a constant force the boundary between bounded and unbounded growth is shifted to higher tubulin concentrations and rescue rates. We determine the critical force f_c which provides the transition from unbounded to bounded growth. (iii) For growth against an elastic obstacle and a non-zero rescue rate, we find a sharply peaked steady state length distribution. The corresponding average length self-organizes such that the average polymerization force equals the critical force f_c for the transition from unbounded to bounded to bounded to bounded to bounded prove that the average polymerization force equals the critical force f_c

BP 25.6 Wed 17:30 Poster C Feedback mechanism for microtubule polymerization regulation — •MARIA ZEITZ and JAN KIERFELD — Physics Department, TU Dortmund, Dortmund, Germany

We investigate the feedback mechanism in the cell between a polymerizing microtubule and the proteins Rac and stathmin and its role in microtubule length regulation. We use two different approaches to investigate the impact of the feedback mechanism: On the one hand, we construct a homogeneous model, which neglects spatial concentration variations and focuses on the feedback effects in the reaction kinetics. This model can be solved analytically. On the other hand, we introduce a more detailed model which couples reactions to diffusion of stathmin proteins and polymerization dynamics of the microtubule. We investigate the the complex dynamics of all three components by stochastic simulations.

BP 25.7 Wed 17:30 Poster C Actomyosin Detection at Fluorescence Interference Contrast Checkpoints for Automated Biocomputation Readout — •MERCY LARD¹, LASSE TEN SIETHOFF², MALIN PERSSON², ALF MÅNSSON², and HEINER LINKE¹ — ¹The Nanometer Structure Consortium (nmC@LU) and Division of Solid State Physics, Lund University, Lund, Sweden — ²School of Natural Sciences, Linnaeus University, Kalmar, Sweden

On-chip biotechnologies, which aim to replace microfluidic, driven flow with active molecular-motor driven transport of cytoskeletal filaments include: bio-computation, diagnostics, and drug screening. These applications would benefit greatly from further miniaturization and increased sensitivity. Here we make use of actomyosin in an in vitro motility assay, incorporated in nanostructures, as a platform for biosimulation of the time evolution of motile objects in complex networks, for example in biocomputation. This type of application can require detection of filaments at checkpoints in the device with high signal-tonoise ratio, for example, to record the number and speed of filaments at a specific location. To serve this need, we make use of fluorescence interference contrast (FLIC) at thin gold lines running perpendicular to nano-sized polymer resist channels that guide filament motion. Using cross-correlation at pairs of Au lines, we strongly enhance S/N and counting accuracy. We demonstrate the tracking of single or multiple filaments at these checkpoints, and discuss limits of this technique. Application areas of this technique will be discussed.

BP 25.8 Wed 17:30 Poster C

Single-molecule motility of kinesin-5 motors between crosslinked microtubules — •ALICE WIESBAUM¹, CHRISTINA THIEDE¹, OLGA ZAITSAVA², VLADIMIR FRIDMAN³, MARCEL JANSON², LARISA GHEBER³, and CHRISTOPH SCHMIDT¹ — ¹Georg-August Universität, Göttingen, DE — ²Wageningen University, Wageningen, NL — ³Ben-Gurion University of Negev, Beer-Sheva, IL

During mitosis, a group of several proteins organizes the dynamics of the mitotic spindle midzone. Two essential functions that need to be fulfilled in the midzone are crosslinking and active sliding of antiparallel microtubules (MTs). In S. cerevisiae, crosslinking is done by the protein Ase1, and sliding is powered by the tetrameric kinesin-5 motors Cin8 and Kip1. It is known that kinesin-5 motors are regulated by binding conditions to MTs. X. laevis Eg5 only moves processively when it is bound between two MTs. Cin8 was found to change its directionality when binding between two MTs. On a single MT, Cin8 is minus-end directed and moves processively. When bound between two MTs, Cin8 drives slow, plus-end directed relative sliding. To test if this property is a cooperative phenomenon of multiple motors or a single-motor property, we performed in vitro TIRF single-molecule experiments. In order to ensure bundling even at low motor concentrations, we employed the crosslinking protein Ase1. We found that single Cin8 motors are capable of switching direction when between MTs. In addition, we analyzed the behavior of a Cin8 mutant, Cin8*tail, which lacks the C-terminal tail domain. This mutant still supports sliding of MTs, but lacks a clear directionality switch.

BP 25.9 Wed 17:30 Poster C

Measurement of F-actin localization tubes and their orientation distribution in three dimensions — •EVELIN JASCHINSKI, INKA KIRCHENBÜCHLER, RONALD SPRINGER, and RUDOLF MERKEL — Forschungszentrum Jülich GmbH, Wilhelm-Johnen-Straße, 52428 Jülich

Under appropriate conditions, the protein actin forms long and semiflexible filaments (F-actin). Due to their large persistence length (~17microns) fluorescently labeled filaments are ideally suited for light microscopic study of conformations and fluctuations. Experiments on labeled test filaments embedded in a non-labeled network enable quantification of the space available, i.e. the well-known localization tube. In recent work our group has analyzed distributions of curvature [1] and diameters of the tubes [2]. These results were analyzed assuming isotropic solutions. However, pronounced and long lasting anisotropy can be induced by sample shear. In order to characterize the impact of directional order of the filaments localization tubes we applied different shear history and measured in three dimension the orientation distributions at different protein concentrations. From them we calculated order parameters and compared the latter to nematic ordering.

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 25.10 Wed 17:30 Poster C

Investigation of desmin intermediate filament assembly by atomic force microscopy — •MAREIKE DIEDING¹, VOLKER WALHORN¹, ANDREAS BRODEHL², HENDRIK MILTING², and DARIO ANSELMETTI¹ — ¹Experimental Biophysics & Applied Nanoscience, Bielefeld University, Germany — ²E. & H. Klessmann Institute for Cardiovascular Research & Development, Heart and Diabetes Centre NRW, Ruhr-University Bochum, Bad Oeynhausen, Germany

Arrythmogenic right ventricular cardiomyopathy (ARVC) is a severe heart muscle disease. It is pathologically characterized by predominant dilatation of the right ventricle and arrhythmias often leading to heart failure or sudden cardiac death. ARVC is often associated with mutations in the desmin intermediate filament (IF) protein. It is known from fluorescence microscopy assays that these mutations can inhibit the formation of extended desmin cytoskeletal networks [1].

We used atomic force microscopy (AFM) topography to study the desmin assembly process *in vitro* at different stages of the filament formation. Thereby we were able to reveal various mutation specific structural defects at distinct stages of the filament assembly. Moreover, our results are nicely supported by complementary methods like cell transfection studies [1]. In future measurements we plan to investigate the assembly process on the single molecule level by AFM single molecule force spectroscopy.

[1] A. Brodehl et al., Dual-color photoactivation localization microscopy of cardiomyopathy associated desmin mutants, J Biol Chem. 287(19), 2012

BP 25.11 Wed 17:30 Poster C Keratin 8/18 Networks and their Interplay with Plectin — •INES MARTIN¹, SOUFI NAFEEY², TOBIAS PAUST¹, MICHAEL BEIL³, HARALD HERRMANN⁴, and OTHMAR MARTI¹ — ¹Department of Experimental Physics, Ulm University, Ulm, Germany — ²Central Facility of Electron Microscopy, Ulm University, Ulm, Germany — ³Clinic of Internal Medicine I, Ulm University, Ulm, Germany — ⁴Division of Molecular Genetics, German Cancer Research Center (DKFZ), Heidelberg, Germany

The keratin 8/18 dimer is a structural building block of intermediate filaments (IFs), which are basic constituents of the cytoskeleton in some epithelial cells. They are responsible for the stiffness of cells and responses to mechanical stimuli. Keratin filaments can be crosslinked by plectin, a protein that links different parts of the cytoskeleton to each other as well as to hemidesmosomes.

In this work we assembled keratin 8/18 and plectin together in vitro to form crosslinked networks. We checked the resulting networks with Scanning Electron Microscopy (SEM) and Immuno-Gold-Labeling and were able to identify the position of plectin molecules. The viscoelastic network properties were measured by passive microrheology and compared to in vitro assembled networks without crosslinker and with MgCl₂ as crosslinker.

BP 25.12 Wed 17:30 Poster C Investigating Intermediate Filament (dis-)assembly Processes with Microfluidics — •BERND NÖDING, VIKTOR SCHRÖDER, JU-DITH BREUER, SUSANNE BAUCH, and SARAH KÖSTER — Institute for X-Ray Physics, University of Göttingen, Germany

The cytoskeleton of eukaryotes consists of three different polymer systems: microtubules, actin filaments and intermediate filaments (IFs). While both microtubules and actin filaments are highly conserved, IFs occur in many different variations. A central property of all filament types is the (dis-)assembly mechanism. For vimentin IFs a principal assembly model exists. However, measurements of the assembly process with high time resolution, which would yield insights especially into the early assembly steps, are still largely missing. To approach this problem, we combine microfluidic and fluorescence techniques in two different ways. First, for studying the fast early assembly steps, we use Fluorescence Cross Correlation Spectroscopy (FCCS) in combination with microfluidic flow channels. With this setup, we will be able to characterize the assembly process with a time resolution in the millisecond range. Second, the later assembly stages and the disassembly of IFs can be observed with a different microfluidic design, which employs reaction chambers coupled via diffusion channels to a very well defined buffer reservoir. Thus the IF assembly and dissassembly process can be influenced by minute changes in the buffer system. Trough the combination of both these methods we aim to form a more complete picture of the assembly mechanism of IFs.

BP 25.13 Wed 17:30 Poster C Passive Microrheology: A model-based approach to determine mechanical properties of assembled networks — •TOBIAS PAUST¹, INES MARTIN¹, MICHAEL BEIL², and OTHMAR MARTI¹ — ¹Institute for Experimental Physics, Ulm University — ²Department of Internal Medicine I, Ulm University

Macro- and microrheology is extensively used to characterize complex networks of biopolymers. A possible way to describe the mechanical properties of a viscoelastic medium is to measure the thermal motion of a particle embedded in the medium and compute the unilateral transformation.

In a model-based approach the mean squared displacement of the particle motion is divided into three terms describing the different properties of the network - elasticity, viscosity and the motion of the particle in a spatial confinement.

The focus of our work are the measurements of in vitro assembled keratin 8/18 networks to underline the functionality of the model-based approach. Furthermore, Brownian motion simulations of spatially confined beads with the influence of the meshes' elasticity and its drift are performed to allow a comparison to the measurements.

BP 25.14 Wed 17:30 Poster C $\,$

Conception and construction of a microfluidic chamber for the optical tweezers — •SAMUEL VOLLMER, TOBIAS PAUST, and OTHMAR MARTI — Institute of Experimental Physics - University Ulm, D-89069 Ulm, Germany

Microfluidics is the science and engineering of systems in which fluid behavior differs from conventional flow theory primarily due to the small length scale of the system. For many measurement systems small length scales are obligatory for the ability to work with small reagent volumes.

It is important to understand the assembly process of Keratin 8/18 networks and their mechanical properties at early times. Therefore we constructed a microfluidic chamber to mix assembly buffer and dialysis buffer at the optical tweezers location to investigate the network mechanics at starting times.

To find the best geometry for our chamber we first have done some simulations using the finite element method. These simulations show the mixing of the two buffers in dependence of the geometry of the chamber.

BP 25.15 Wed 17:30 Poster C $\,$

Laser ablation on actomyosin network — •ARNAB SAHA¹, GUIL-LAUME SALBREUX¹, FRANK JÜLICHER¹, and STEPHAN GRILL^{1,2} — ¹Max Planck Institute for the Physics of Complex Systems Nöthnitzer Straße 38 01187 Dresden Germany — ²Max Planck Institute of Molecular Cell Biology and Genetics Pfotenhauerstr. 108 01307 Dresden, Germany

The cell cortex is a thin layer beneath the membrane that largely consists of cross-linked actin filaments, non-muscle myosin motor protein and a plethora of actin binding proteins (ABPs). Collective dynamics of myosin over actin meshwork (assembled by ABPs), that converts chemical energy to mechanical work by ATP hydrolysis, generates active contractility at large cellular length scales. Contractility can build up mechanical stress in the cortex. Sudden up-or-down regulation of myosin induces spatial inhomogeneity in the stress profile which finally leads to cortical flow.

One need to investigate the flow pattern in order to understand the physical properties of the actomyosin network. Here we describe the actomyosin mesh at a coarse grained level as a viscoelastic, active and contractile gel. We develop a two dimensional hydrodynamic model, valid at long length scales and short to long time scales (incorporating both elastic and viscous regimes), using basic symmetry principles and conservation laws. The description is then used to investigate the spatio-temporal dynamics observed after the laser ablation on actomyosin cable during zebrafish epiboly.

BP 25.16 Wed 17:30 Poster C

Correlating the signaling cascade with movement in Dictyostelium discoideum — •CHRISTOPH BLUM^{1,2}, VLADIMIR ZYKOV^{1,2}, KAUMUDI PRABHAKARA^{1,3}, MARCO TARANTOLA^{1,4}, and EBERHARD BODENSCHATZ^{1,2,3} — ¹Max-Planck-Institu für Dynamik und Selbstorganisation, Göttingen, Germany — ²Georg-August-Universität, Institut für Nichtlineare Dynamik, Göttingen, Germany — ³Cornell University, Ithaca, USA — ⁴University of California, San Diego, USA

Actin cytoskeletal dynamics provide the fundamental basis of eukaryotic cell motility. The cross-linked actin network at the front of a cell pushes the leading edge of the membrane towards the source of attractant. It is our aim to provide a quantitative understanding of the spatio-temporal dynamics of the actin cytoskeleton within the actin cortex.

We have developed experimental methods to address single Dictyostelium discoideum cells with well-controlled mechanical and chemical stimuli. Our experimental techniques are based on microfluidic devices, such as flattening device and micromixer, and fluorescence microscopy (Confocal, TIRF).

Here we present correlations of an important protein in the signaling cascade(Ras-GTP) with the actin polymerization as well as correlations of Ras-GTP localization with pseudopod dynamics. The localization is visualized by the Ras binding domain(RBD-GFP). The pseudopod formation is analyzed by curvature maps. The polymerization of actin is shown by the filametous actin marker LimE.

BP 25.17 Wed 17:30 Poster C **Rheology of cytoskeletal in-vitro networks** — •MEENAKSHI PRABHUNE¹, KNUT HEIDEMANN², FLORIAN REHFELDT¹, MAX WARDETZKY², and CHRISTOPH SCHMIDT¹ — ¹Third Physics Institute, Georg August University, Göttingen — ²Department for Numerical and Applied Mathematics, Georg August University, Göttingen

Living cells are governed by active cellular processes such as cell division, adhesion and migration that depend on the cytoskeleton. The cytoskeleton is a composite cross-linked polymer network of cytoskeletal filaments ranging from rod-like microtubules and actin bundles to semi-flexible actin filaments and softer intermediate filaments. Singlecomponent in vitro networks have been studied, but well defined composites are more difficult to construct and are not yet well understood. Here, we have generated heterogeneous networks in vitro by cross-linking microtubules and ds DNA via a heterobifunctional crosslinker (sulpho SMCC). DNA as a cross-linker has the unique advantage of having a well-defined length, which we vary in our experiments. We have measured the shear-elastic response in these networks by macrorheology experiments at varying strains and frequencies. The nonlinear response was also characterized using differential stiffness measurements in a macrorheometer. Simultaneously, we compare the experimental data to numerical simulations that we have developed for networks of stiff slender rods connected by semi-flexible linkers (see poster by Knut Heidemann).

BP 25.18 Wed 17:30 Poster C Biopolymer Networks: Simulations of Rigid Rods Connected by Wormlike Chains — •KNUT M HEIDEMANN¹, MEENAKSHI M PRABHUNE², FLORIAN REHFELDT², CHRISTOPH F SCHMIDT², and MAX WARDETZKY¹ — ¹Institut für Numerische und Angewandte Mathematik, Georg-August-Universität Göttingen — ²Drittes Physikalisches Institut - Biophysik, Georg-August-Universität-Göttingen

The cytoskeleton of cells is a composite network of filaments ranging from stiff rod-like microtubules to semiflexible actin filaments that together play a crucial role in cell structure and mechanics. The collective dynamics of these cytoskeletal filaments with different mechanical properties are yet to be understood completely. To model such a strongly heterogeneous composite, we simulate networks of *rigid* rods connected by *flexible* linkers (wormlike chains). We extract elastic moduli by quasistatic deformations at varying filament densities and analyze the crossover between cross-link dominated and rod dominated regimes. In particular, we are interested in the asymptotic stress dependence of the *differential modulus*.

The simulations are accompanied by rheological experiments on networks of *microtubules* (MTs) cross-linked by double-stranded DNA of variable length (cf. poster Meenakshi Prabhune).

BP 25.19 Wed 17:30 Poster C Actin Dynamics in Myosin II-null Dictyostelium Discoideum — HSIN-FANG HSU^{1,4}, •CHRISTIAN WESTENDORF^{1,4}, CARSTEN BETA², and EBERHARD BODENSCHATZ^{1,3,4} — ¹Max Planck Institute for Dynamics and Self-Organization, Goettingen, Germany — ²Institute of Physics and Astronomy, University of Potsdam, Potsdam, Germany — ³Laboratory of Atomic and Solid-State Physics, Cornell University, USA — ⁴Institute for Nonlinear Dynamics, Georg-August University, Goettingen, Germany

Starved D. discoideum show chemotaxis to cAMP. The directed migration is driven by the assembly and disassembly of actin filaments. In addition to the actin dynamics the contraction of Myosin II plays an important role in the effective movement. Cells lacking Myosin II show frequent protrusions of the membrane. Here we investigate the dynamics of actin filament (labeled by LimE-GFP) in the AX2 cells without Myosin II heavy chain. We observed Myosin II mutants show spontaneous periodic actin dynamics even without external stimulation of cAMP. We further investigate this phenomenon by perturbing the system with different periodic pulses of cAMP created by flow photolysis. Our observation showed Myosin mutants show more instability of actin networks than wild-type cells.

BP 25.20 Wed 17:30 Poster C Active Biopolymer Networks — •Adam Walker, Nikta Fakhri, Kerstin von Roden, and Christoph F. Schmidt — Drittes Physikalisches Institut - Biophysik, Georg-August-Universität, Göt-

tingen

Biological cells rely on the cytoskeleton - an active, polymeric structure - to drive essential dynamic processes such as motility, growth and cell division. In contrast to technical materials, a living cell is a non-equilibrium system. This research focuses on the investigation of mechanical properties and dynamics of model biopolymer networks quantitatively using a largely experimental approach centered on both bulk rheology and optical-trapping based microrheology. Non-muscle Myosin IIa motors were characterized using fluorescence microscopy for in vitro actin motility assays and shown to interact with and translocate actin filaments. Initial macro-rheology experiments dealt with model networks constructed purely from actin filaments and allowed us to quantify both the linear- and nonlinear- response of the networks. Networks will be activated using myosin motors and associated crosslinking proteins to better approximate the real cytoskeletal environment.

 SCHELL, and CLAUDIA VEIGEL — Division of Cellular Physiology and CeNS, LMU München, Schillerstrasse 44, 80336 München

The genome of the protozoan Leishmania parasite comprises just two classes of myosin, however only class XXI was shown to be expressed in both stages of the parasites life cycle. Apparently the only myosin motor in Leishmania, myosin-XXI is a unique model system and thought to perform a large variety of functions ranging from membrane anchorage to long-range directed cargo movement. However, nothing is known about the oligomerisation states, motility or cargo-binding of this motor. Using size exclusion chromatography, motility assays and electron microscopy we found that, in the absence of calmodulin, the motor formed immobile dimers. In the presence of calmodulin the motor was monomeric and moved actin filaments at ~15 nm.s-1. The monomers bound to liposomes, while the dimers did not. We identified phospholipid binding domains that overlapped with the dimerisation domains, including a PX-domain overlapping with the converter region. We propose a novel mechanism of myosin regulation where myosin-XXI monomers bind to lipid cargo and act as transporters, while the dimeric form is unable to bind to lipids and acts as an immobile, ATP-dependent crosslinker of actin filaments. Supported by SFB 863, CeNS, Baur-Stiftung