BP 21: Biopolymers and Biomaterials II (with CPP)

Time: Wednesday 10:15–13:00 Location: ZEU 260

Invited Talk BP 21.1 Wed 10:15 ZEU 260

Stretching Proteins out of equilibrium: how extracellular matrix proteins serve as mechanotransducers — •Viola Vogel — Department of Materials, ETH Zürich, Switzerland

While physical factors and material properties regulate many cell functions, the underpinning mechanisms how cells and tissues sense mechanical stimuli and convert them into biochemical signals are not well understood. As cells explore their environments, they pull on extracellular matrix and thereby stretch those proteins that physically connect the exterior microenvironment with the contractile cytoskeleton. Detailed mechanisms will be discussed how the stretching of proteins can switch their functional display. Deciphering how proteins can serve as mechano-chemical signaling switches is not only essential to learn how cells probe and respond to their environments, but it has also far reaching implications in tissue engineering, systems biology and medicine.

BP 21.2 Wed 10:45 ZEU 260

Formation and Confinement of Actin Networks in Microchambers — •Siddharth Deshpande[1], Dagmar Steinhauser[2], and Thomas Pfohl[1,2] — [1]Chemistry Department, University of Basel, Switzerland — [2]Max Plank Institute for Dynamics and Self Organization, Göttingen, Germany

Our aim is to study the spatiotemporal evolution of biopolymer networks (e.g. actin, collagen, fibrin) with the aid of microfluidics and using a bottom-up approach. We have designed microfluidic devices consisting of microchambers of different shapes and sizes connected to the main channel by narrow connecting channels. High flow conditions can be achieved in the main channel to control the concentration and composition of the aqueous solution while the transport of molecules into the microchambers is governed by diffusion.

Rhodamine labeled actin monomers are used for the experiments and visualized by fluorescence microscopy. Once polymerized, the actin filaments formed inside the chamber are confined and form an entangled actin network, which can be analyzed for various network properties such as connectivity distribution of nodes, length distribution of links, node fluctuations, link fluctuations and fluctuations in the mesh size.

The experiments with actin bundles in confinement show that the persistence length of actin bundles (L_p) increases proportionally with the number of filaments present in a bundle (n) as: $L_p \approx n^{1.3}$. In the next step, we try to form more complex networks using cross-linking proteins such as α-actinin, filamin, HMM and use FRET microscopy to analyze it.

BP 21.3 Wed 11:00 ZEU 260

Mechanics and Dynamics of Individual Intermediate Filaments — •Bernd Nöding, Susanne Bauch, and Sarah Köster — Courant Research Centre Nano-Spectroscopy and X-Ray Imaging, University of Göttingen, Germany

The mechanical rigidity of a polymer is characterized by its persistence length L_P. In the case of the intermediate filament (IF) protein vimentin, L_P was found to be on the order of one micrometer using static measurement methods. In contrast, we perform dynamic measurements on fluorescently labeled IFs confined in microchannels, thereby realizing the Odijk confinement regime. Since IFs can be classified as semiflexible polymers ($L \approx L_P$) we assume the worm-like chain model for our fluctuation analysis. The channel walls are included as a parabolic potential in our calculations. Interaction of the filament and the confining microchannel gives rise to an additional length scale, the deflection length λ. We combine IF data with literature data for actin. Thereby we can access both the channel dimension d and L_P, which define the scaling law connecting λ and L_P, $\lambda = a \cdot d^{2/3} \cdot L_P^{1/3}$. The scaling law is fully confirmed by our experiments. Additionally our dynamic measurements yield L_P on the order of one micrometer for vimentin filaments.

BP 21.4 Wed 11:15 ZEU 260

A constitutive law for cross-linked actin networks by homogenization techniques — Denis Caillerie[1], •Karin John[2], Chaouqi Misbah[2], Philippe Peyla[2], and Annie Raoult[3] — [1]L3S-R, BP 53 - 38041 Grenoble Cedex 9, France — [2]LSP, UJF Grenoble & CNRS, BP 87 - 38402 Saint-Martin-d'Hères, France — [3]LMAP5, Université Paris Descartes, 45 rue des Saints Pères, 75270 Paris Cedex 06, France

Inspired by experiments on the actin driven propulsion of micrometer sized beads we develop and study a minimal mechanical model of a two-dimensional network of stiff elastic filaments grown from the surface of a solid circle. Starting out from a discrete model of the network structure and of its microscopic mechanical behavior we derive a macroscopic constitutive law by homogenization techniques. We calculate the axisymmetric equilibrium state and study its linear stability depending on the microscopic mechanical properties. We find that thin networks are linearly stable, whereas thick networks are unstable. The critical thickness for the change in stability depends on the microscopic elastic constants. The instability is induced by the increase in the compressive load on the inner network layers as the thickness of the network increases. The here employed homogenization approach combined with more elaborate microscopic models can serve as a basis to study the evolution of polymerizing actin networks and the mechanism of actin driven motion.

15 min. break

BP 21.5 Wed 11:45 ZEU 260

Dynamics and mechanics of formin mediated actin bundles — •Florian Rückerl, Timo Betz, and Cécile Sykes — UMR168, Institut Curie, Paris

In our experiments actin filaments and actin bundles are produced by polymerization by the formin mDia1(FH1FH2). To probe their dynamics and mechanics, we use a state of the art optical tweezers setup and create multiple traps (2 to 5) with acousto-optical deflectors (AODs). Digitally controlled AODs in time sharing mode allow to position and move several traps simultaneously. Employing a four quadrant diode as a position detector results in high temporal and spatial resolution, 10μs and <1nm, respectively. This allows to investigate the polymerizing dynamics of mDia1 by directly observing the deflection of formin coated beads inside the optical trap. Preliminary results indicate single monomer addition events at infrequent intervals.

By attaching several beads to individual bundles we can create pico-Newton forces in arbitrary directions. The mechanical properties of the bundle are then probed by bending, pushing and pulling on the bundle. We find that pulling on a bundle leads to its elongation, presumably by relative sliding of the bundle filaments to each other.

Furthermore, the setup can be used for the direct manipulation of the bundles without beads attached to it, allowing for an *in situ* non invasive measurement. Correlating the local fluctuations at several positions on the bundle yields its persistence length and gives an estimate of the number of filaments in the bundle.

BP 21.6 Wed 12:00 ZEU 260

Network Formation of Cytoskeletal Proteins — •Christian Dammann, Bernd Nöding, Susanne Bauch, and Sarah Köster — Courant Research Centre Nano-Spectroscopy and X-Ray Imaging, University of Göttingen, Germany

The structure and function of biological systems is determined by their bio-environment. Therefore, a drop-based microfluidic device is tailored to probe context-sensitivity of biological systems. In this device a series of monodisperse aqueous drops is created and used as picoliter bio-compartments. The composition of the drops is varied from drop to drop. Thus, the biological system is encapsulated in drops with tunable chemical content. These drops are then stored in the device for long-time observations. The content composition of each individual drop can be reconstructed. Possible applications of this tool are manifold. The device proves to be suitable for *in vitro* studies on cytoskeletal proteins. We focus on the assembly and network formation of vimentin intermediate filament. The assembly of vimentin depends on the ionic strength. We are able to directly image the networks of the fluorescently tagged protein and show that divalent ions induce compaction of these networks.

BP 21.7 Wed 12:15 ZEU 260

Functionalized lipid bilayers for rapid cell attachment — •Samira Hertrich, Martin Huth, and Bert Nickel — Ludwig-Maximilians-Universität, Department für Physik and CeNS,

Geschwister-Scholl-Platz 1, 80539 München

The behaviour of cells in contact to interfaces varies significantly depending on the surface properties. Bioadhesive coatings can act as an interlayer between cells and anorganic interfaces tuning the interaction of cells with the surface. Here, a multilayer system consisting of a supported lipid bilayer and two protein layers is used to produce a surface favoring neural stem cell attachment. Biotin and streptavidin act as a layer of linkers in between the lipid bilayer and the cell adhesive polypeptide AK-cyclo[RGDfC].

The trilayer system was characterised by x-ray reflectometry (D4, HASYLAB) and neutron reflectometry (N-REX, FRM2) measurements, which allowed for the determination of the layers thicknesses and the hydration of both lipids and proteins. Cell attachment to the coated surface was verified via fluorescence microscopy [1]. Microscopy confirms rapid attachment of stem cells while reflectometry indicated a dense on edge configuration of the AK-cyclo[RGDfC] thus maximizing the number of exposed RGD groups. Experiments have been performed in collaboration with E. Madarasz and G. Menzo from the Hungarian Academy of Science (HAS).

[1] Huth, M, et al., Materials 2010, 3, 4994-5006.

BP 21.8 Wed 12:30 ZEU 260

Two-component Polymer Scaffolds for Controlled Three-dimensional Cell Culture — •Benjamin Richter[1,2], Franziska Klein[1], Thomas Striebel[1], Clemens Franz[1], Georg von Freymann[3], Martin Wegener[2], and Martin Bastmeyer[1] — [1]Zoologisches Institut, Karlsruher Institut für Technologie, 76131 Karlsruhe — [2]Angewandte Physik, Karlsruher Institut für Technologie, 76131 Karlsruhe — [3]AG Optische Technologien und Photonik, Technische Universität Kaiserslautern, 67663 Kaiserslautern

Fibrous collagen or matrigel matrices are commonly used to study three-dimensional (3D) cell behaviour, but these matrices have a random pore size and are structurally and chemically ill defined. We and others have recently shown that direct laser writing (DLW) is a versatile technique to fabricate tailored 3D cell-culture scaffolds in the micrometer to nanometer range. By using an adequate photoresist, elastic 3D scaffolds for cell-force measurements have also been realized. These DLW scaffolds have been homogeneously coated with ECM molecules. Ideally, they should rather have an adjustable distribution of cell-substrate contact sites to manipulate cell adhesion and cell shape in all three dimensions. By sequential DLW of two different photoresists, composite-polymer scaffolds with distinct protein-binding properties are fabricated and selectively bio-functionalised thereafter. Cells cultured in these scaffolds selectively form cell-adhesion sites with the functionalised parts, allowing for controlling cell adhesion and cell shape in 3D - forming the basis for future designer tissue-culture scaffolds.

BP 21.9 Wed 12:45 ZEU 260

Characterizing bacterial adhesion: The role of van der Waals forces — •Nicolas Thewes, Peter Loskill, Sebastian Hümbert, and Karin Jacobs — Deparment of Experimental Physics, Saarland University, 66041 Saarbrücken, Germany

Bacterial adhesion to surfaces is a complicated process that not only depends on the type of bacterium and the type of surface, but also on subsurface composition, as we have shown in a recent study. To probe the adhesion of *s. carnosus*, various surfaces have been prepared, ranging from hydrophilic to hydrophobic, from smooth to rough surfaces. To probe the effect of subsurface composition on the adhesion strength, Si wafers with different Si oxide layer thicknesses have been used. Clearly, the adhesion is stronger on wafers with thin Si oxide layer, irrespective if the wafer was hydrophobized by a monolayer of silanes or not, which is a clear evidence that long-range van der Waals forces play a crucial role for bacterial adhesion. It moreover shows that subsurface composition must be taken as characteristics of a sample, much in the same way chemical composition, wetting properties or surface roughness are taken into account. An additional parameter to control is the proper immobilization of the bacteria on the AFM tip, with the help of which force/distance curves have been performed.