BP 23: Cell Motility and Migration (in vitro and in vivo)

Time: Thursday 14:00–17:00 Location: H43

Invited Talk BP 23.1 Thu 14:00 H43

The Physics of Neuronal Growth — •Timo Betz, Daniel Koch, and Josef Käs — Institut for Soft Matter Physics, University of Leipzig, Germany

The correct development of the central nervous system requires accurate and reliable neuronal network formation, a process accomplished by a highly dynamic structure at the tip of a growing neurite, called the growth cone. To find its proper target, each growth cone integrates chemical and mechanical signals, and converts these signals into changes of its active polymeric cytoskeleton. To understand these fundamental growth processes, we quantified the physical properties of a growth cone, like traction forces, viscoelastic properties and actin dynamics, and found that growth cones show unique features with respect to other motile cell types.

We show that bistable stochastic fluctuations between growth and retraction phases determine the direction of neuronal growth, and that these fluctuations are dominated by bistable modes of actin polymerization. Furthermore, the viscoelastic properties of the growth cone are combined with the measured actin dynamics to calculate the active internal and external forces generated by the growth cone. Integrating these data suggests that growth cones utilize physical processes like stochastic signal amplification and unique mechanical properties to build an organism's intricate nervous system. Moreover, our measurements provide the base to understand the complex interplay between actin polymerization, active internal forces and substrate adhesion that finally results in nerve regeneration and neuronal network formation.

BP 23.2 Thu 14:30 H43

Origins and Limitations of Optical Neuron Guidance — •Daniel Koch, Timo Betz, Allen Ehrlicher, Björn Stuhrmann, Michael Gögler, and Josef Käs — Institute for Soft Matter Physics, University of Leipzig, Linnestr. 5, 04103 Leipzig, Germany

The growth cone, a highly motile sensory structure at the tip of an advancing neurite, plays a fundamental role in the wiring of neuronal connections during development, nerve regeneration, and in neuronal plasticity. The dynamics of the leading edge is governed by actin polymerization and can be described as a bistable stochastic process which means polymerization is either turned on or off. Furthermore, the direction taken by the growth cone is introduced by the orientation of the filopodia which are force sensitive actin filament bundled structures that emerge from the leading edge. In actively extending growth cones, a laser spot placed at the leading edge affects the direction taken by the growth cone. However, optical control is not achieved in non-extending growth cones with polymerization mostly turned off which sets a natural limit for optical neuron guidance. The analysis of optically induced turning events reveals that the applied optical forces lead to filopodia reorientation. We hypothesize that the filopodia are coupled back into the central region and in a lever arm like fashion change the extension of the leading edge.

BP 23.3 Thu 14:45 H43

Glial Cell Stiffness as Guidance Cue for Neurons — •Kristian Franze[1,2], Timo Betz[1], Yunbi Lu[1,2], Johannes Bayer[3], Melike Lakadamyali[3], Paul Janmey[4], and Josef Käs[1] — [1]Soft Matter Physics, Universität Leipzig — [2]Paul-Flechsig-Institute of Brain Research, Universität Leipzig — [3]CNLD, University of Texas, Austin, USA — [4]Inst. Medicin & Engineering, University of Pennsylvania, Philadelphia, USA

Neuronal migration is a fundamental event during development. Neurons travel from the ventricular zone, the place of their origin, to the cortical plate, bridging distances that can be a multiple of their length. Radial glial cells, which are cells that connect the ventricular zone with the opposing cortical surface with two long, radial processes, are known to guide neuronal migration. Neurons attach to these cells and precisely follow their processes, even if they are significantly bent. No biochemical guidance cues have been identified for this behavior and simple diffusive gradients cannot explain how neurons follow the bent glial shape. We found that in vitro neurons actively probe their mechanical environment. They retracted their processes and reextended them in a random direction when mechanical stresses exceeding ˜300 Pa opposed their leading edge. This threshold corresponds to the maximum substrate stiffness that neurons could visibly deform. Interestingly, radial glial cells were softer than 300 Pa, suggesting that their mechanical properties may facilitate neuronal radial migration in the developing brain. This is in sharp contrast to the current opinion that neuronal guidance is solely based on biochemical signaling.

BP 23.4 Thu 15:00 H43

Symmetry breaking in actin gels - Implications for cellular motility — •Karin John, Philippe Peyla, and Chaouqi Misbah — Université Joseph Fourier Grenoble, Laboratoire de Spectrométrie Physique, BP 87 - 38402 St.-Martin-d'Hères, France

The physical origin of cell motility is not fully understood.

Recently minimal model systems have shown, that polymerizing actin itself can produce a motile force, without the help of motor proteins. Pathogens like Shigella or Listeria use actin to propel themselves forward in their host cell.

The same process can be mimicked with polystyrene beads covered with the activating protein ActA, which reside in a solution containing actin monomers. ActA induces the growth of an actin gel at the bead surface. Initially the gel grows symmetrically around the bead until a critical size is reached. Subsequently one observes a symmetry breaking and the gel starts to grow asymmetrically around the bead developing a tail of actin at one side. This symmetry breaking is accompanied by a directed movement of the bead, with the actin tail trailing behind the bead. Force generation relies on the combination of two properties: growth and elasticity of the actin gel.

We study this phenomenon theoretically within the framework of a linear elasticity theory and linear flux-force relationships for the evolution of an elastic gel around a hard sphere.

Conditions for a parity symmetry breaking are identified analytically and illustrated numerically with the help of a phasefield model.

BP 23.5 Thu 15:15 H43

A Biomimetic System Modeling Active Lamellipodial Network Dynamics — •Florian Huber, Björn Stuhrmann, and Josef Käs — Institute for Soft Matter Physics, University Leipzig, Linnéstr. 5, D-04103 Leipzig, Germany

Many different cell types (e.g., keratocytes or fibroblasts) show directed motion (motility), a process driven by the assembly of Actin protein at the leading front of the cell, the lamellipodium. Although the details of polymerization regulation by accessory proteins differ between cell types, several important features are conserved throughout the eukaryotic kingdom. The key molecular players involved in these processes have been identified and have already been used to generate *in vitro* Actin network growth. While existing assays are sufficient to explain intracellular bacteria propulsion, they are not adequate to describe crawling cell motility extensively. The required next step towards cellular conditions is to confine the polymerizing Actin gel to nanostructured cell-sized chambers. This approach restricts the protein pool available and thus allows to mimic for the first time the self-sustaining character of the lamellipodia machinery.

Our setup will allow the observation of the system's response to controlled variation of various biochemical and physical parameters. Numerical simulations will be used to extract constitutive equations from experimantal data on dynamic distributions of the various protein species. Mechanical properties will be analyzed using passive microrheology. This model system represents a novel means to explore biomechanical mechanisms forming the basis of cell motility.

BP 23.6 Thu 15:30 H43

Investigation of Filopodial Mechanics and Dynamics — •Brian Gentry[1], Michel Gögler[1], Marie-France Carlier[2], and Josef Käs[1] — [1]Universität Leipzig, Linnestr. 5, Leipzig, Germany — [2]CNRS, LEBS, 1 Avenue de la Terrasse, Gif-sur-Yvette, France

The actin cytoskeleton is a complex system that dynamically reorganizes its structure to produce forces that drive the leading edge of a cell membrane outward. Filopodia emerge from local reorganization of the dense lamellipodial filament network. Stiff bundles are essential to produce protrusive forces, so we study their mechanical properties. We directly measure actin bundle bending stiffness in vitro, providing information about crosslinker and bundle characteristics. Formation of long, unbranched bundles also requires that fiber ends be protected from capping. Formin is an end-binding molecule which is capable of

nucleating and driving the polymerization of actin polymers in vivo, remaining bound simultaneously to both fiber and substrate. We are studying formin in a controlled, reconstituted system to help elucidate its precise functions as considered in recent models. Both experiments use a state-of-the-art laser tweezer to track the position of a bead attached to an actin filament bundle, allowing us to measure critical buckling and motor-driven forces. The formin measurements are the first for an end-tracking motor and will lend new insight into the underlying dynamics of its operation. Our setup allows us to take a novel approach to the study of filopodial component's properties-bundles which provide stiffness and a molecular engine which produces adequate forces such that protrusion can occur.

BP 23.7 Thu 15:45 H43

Dynamics of receptor-ligand binding and filopodial retraction in phagocytosis — •Alexander Rohrbach[1] and Holger Kress[2,3] — [1]University of Freiburg, Germany — [2]EMBL, Heidelberg, Germany — [3]present address: Yale University, New Haven, USA

Phagocytosis is the process by which bacteria are internalized into macrophages. This process, which is a central mechanism in the immune system, was so far mainly investigated by conventional light and electron microscopies. However, its mechanical properties were barely known up to now. We used optical tweezers-based microscopy to investigate the mechanics of phagocytosis. The motion of an optically trapped bead was tracked interferometrically in 3D with nanometer precision at a microsecond timescale.

The measurement of the thermal bead fluctuations during the binding to the cell membrane enabled the observation of individual receptor-ligand bond formation. These observations were in agreement with Brownian dynamics simulations of the binding process. By inducing binding of beads to filopodia, we found that filopodia act as cellular tentacles: They retract a few seconds after binding and pull the bound beads towards the cell. The observation of discrete F-actin dependent 33-nanometer steps during retraction led to the hypothesis that an actin-based molecular motor plays an important role in the retraction. Force-velocity measurements revealed the mechanical properties of this putative motor. A model for the force-dependent motor kinetics confirming these results was developed.

BP 23.8 Thu 16:00 H43

Keratocyte migration as active Brownian motion: Experiments and theory — •Simon Flyvbjerg Tolic-Norrelykke and Frank Julicher — Max Planck Institute for the Physics of Complex Systems, Dresden, Germany

Fish keratocytes constitute a popular model system for the study of cell migration. Keratocytes are highly mobile cells that live on the outside of fish-scales where they are thought to be involved in wound healing and repair.

We will present some recent experimental findings and theoretical results for the migration of individual keratocytes in two dimensions. Cells were tracked as they migrated on a glass surface in the absence of external stimuli and parameters were extracted from the time-series of positions, allowing for the construction of an equation of motion. In contrast to other existing models for migration, these cells turned out to be best described in terms of an active Brownian process. In active Brownian processes internal energy is converted into motion, leading to a preferred speed that is different from zero and velocity histograms with a characteristic donut shape. Analytical expressions for observable parameters are derived and compared to the experimental data as well as to results from simple simulations.

BP 23.9 Thu 16:15 H43

driving forces in cell motility, motors versus polymerization — •Claudia Brunner, Michael Gögler, Allen Ehrlicher, and Josef Käs — Universität Leipzig, Linnestr 5, 04103 Leipzig

A cell's ability to move is fundamental for various functions in nature, such as morphogenesis, immune response, and the invasiveness of cancer. On the molecular level, actin polymerization and molecular motors, such as myosin, are involved in cell motility but the mechanism as a whole is not very well understood. Here we present direct measurements of the forward forces generated at the leading edge of the lamellipodium and at the cell body of rapidly translocating fish keratocytes. Our SFM-based technique uses the vertical and lateral deflection of the cantilever to directly measure the maximal forward force of whole cells by stalling them. Through selective manipulation of molecular components by addition of different drugs, the stall forces and the velocity correlation can be compared to elucidate the importance of different force generating processes, such as polymerization and molecular motors.

BP 23.10 Thu 16:30 H43

Role of viscosity and surface tension of zebrafish embryonic tissues in tissue flows during gastrulation — •Eva-Maria Schoetz[1,2], Tigran Bacarian[3], Malcom Steinberg[4], William Bialek[4], Carl-Philipp Heisenberg[1], Ramsey Foty[5], and Frank Julicher[2] — [1]MPI-CBG, Dresden — [2]MPI-PKS, Dresden — [3]UCI, USA — [4]Princeton University, USA — [5]UMDNJ, USA

At the onset of gastrulation in zebrafish, complex flows and cell movements occur, which are not well understood. Here, we study the material properties of zebrafish embryonic tissues which are important for the tissue dynamics. We found that these tissues behave viscoelastic and exhibit liquid-like properties on long time scales. They relax internal stress caused by compressive forces or, in the absence of external forces, round up and fuse into spheres to minimize their free surface. Quantitative differences in the adhesivity between different types of tissues result in their immiscibility and sorting behavior analogous to that of ordinary immiscible liquids. When mixed, cells segregate into discrete phases, and the position adopted correlates with differences in the aggregate surface tensions for these phases. Surface tensions were measured with a tissue surface tensiometer. Aggregates were compressed and their force response and shape were recorded as a function of time. From the analysis of the force-relaxation curves, we determined the surface tensions, relaxation times, tissue viscosities and shear moduli. Furthermore, by 4D-cell tracking, we measured kinetic parameters such as cell speed, directionality and persistence of cell movement.

BP 23.11 Thu 16:45 H43

Continuum Description of Growing Cellular Tissues — •Thomas Bittig[1], Ortrud Wartlick[2], Anna Kicheva[2], Marcos González-Gaitán[2], and Frank Jülicher[1] — [1]Max Planck Institute for the Physics of Complex Systems, Nöthnitzer Str. 28, 01187 Dresden, Germany — [2]Department of Biochemistry and Department of Molecular Biology, Geneva University, Sciences II, Quai Ernest-Ansermet 30, 1211 Geneva 4, Switzerland

During the development of multicellular organisms, organs grow to well-defined shapes and sizes. The mechanisms that coordinate the proliferation and movement of cells in growing tissues remain still unclear. In order to study cellular movement in a growing epithelium, we developed a continuum description which considers the time evolution of a local cell density in two or three dimensions. We describe the tissue as a viscous fluid in which active stresses are generated by cell division. We consider situations where cell division is randomly oriented and where a preferred orientation of cell division exists. We perform numerical studies of this macroscopic description using a discrete model on a cellular level. Our descriptions can be used as a basis for the study of the transport of signaling molecules through growing tissues as e.g. in the growing Drosophila wing disc, a precursor of the fly wing.