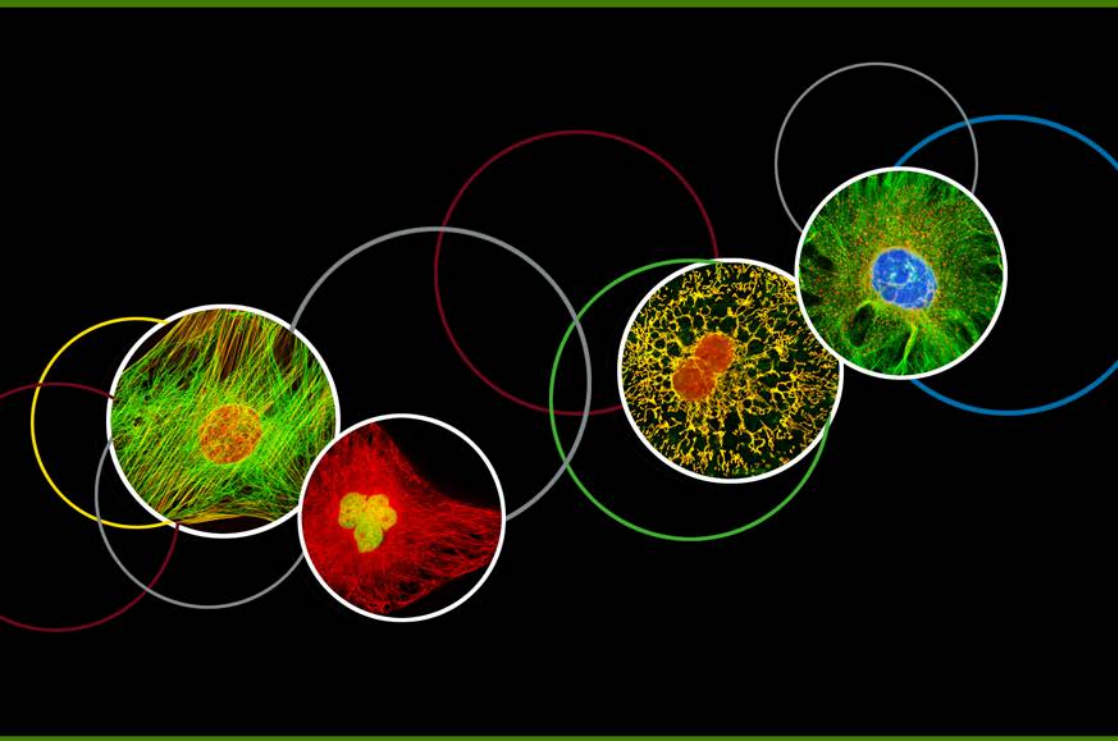
BOOK OF ABSTRACTS



Cell Physics 2014

23.-26. September 2014
Saarbrücken

SFB 1027

GRK 1276

DFG

UNIVERSITÄT DES SAARLANDES

Welcome…

…to the conference "Cell Physics 2014" at the Saarland University in Saarbrücken, Germany, 23.-26.9.2014. The conference is organized and financed by the Collaborative Research Center *SFB 1027* "Physical modeling of non-equilibrium processes in biological systems" and the Graduate School *GRK 1276* "Structure formation and transport in complex systems". It is intended to be an interdisciplinary platform for scientific exchange between participants from cell biology and biophysics, both represented in roughly equal numbers, and focusses centrally on theoretical concepts in conjunction with cell biological experiments. Topics include

- Cellular Self-Organization
- Collective Processes in Cells
- Cytoskeleton Physics
- Molecular Motors
- Intracellular Transport
- Calcium Signaling
- Cellular Communication

The official conference will start Tuesday, 23.9.2014 at 8:50 am, and finish around lunchtime Friday, 26.9.2014. It consists of invited talks, contributed oral presentations and poster sessions. A pre-conference workshop for the members of the SFB 1027 and the GRK 1276 will take place on Monday, 22.9.2014. All participants of the Cell Physics conference are cordially invited to join this pre-conference event comprising oral presentations and many posters of the SFB 1027 and GRK 1276.

Heiko Rieger, Ludger Santen, and Manfred Lücke
(Saarland University, Germany)

Table of Contents

UNIVERSITÄT DES SAARLANDES

Cell Physics 2014

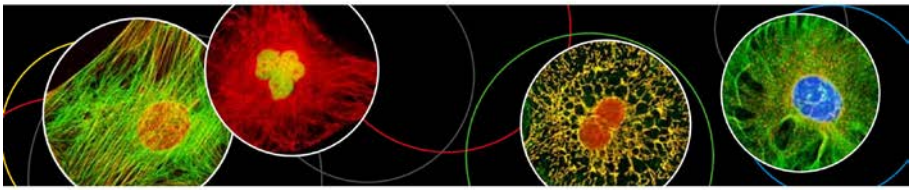
23.-26. September | Saarbrücken

TOPICS

Cellular Selforganization
Collective Processes in Cells
Cytoskeleton Physics
Molecular Motors
Intracellular Transport
Calcium Signalling
Cellular Communication

CONFIRMED SPEAKERS

Roy Bar-Ziv (Weizmann Institute of Sciences, Israel)
Sonia Cortassa (Johns Hopkins School of Medicine, Baltimore, USA)
Steve Gross (University of California, Irvine, USA)
Vincent Hakim (Ecole Normale Supérieure, Paris, France)
Lars Hufnagel (EMBL, Heidelberg, Deutschland)
Jean-Francois Joanny (Institut Curie, Paris, France)
Paul Janmey (University of Pennsilvania, Philadelphia, USA)
Frank Jülicher (Max-Planck-Institut, Dresden, Deutschland)
Josef Käs (Universität Leipzig, Deutschland)
Richard Lewis (Stanford University School of Medicine, USA)
Reinhard Lipowsky (Max-Planck-Institut, Potsdam, Deutschland)
Nicolas Minc (Institut Jacques Monod, Paris, France)
Dan Needleman (Harvard SEAS, Cambridge, USA)
Jose Onuchic (Rice University, Houston, USA)
Ian Parker (University of California, Irvine, USA)
Jacques Prost (Institut Curie, Paris, France)
Christoph Schmidt (Universität Göttingen, Deutschland)
Roland Wedlich-Söldner (Universität Münster, Deutschland)
David Yule (University of Rochester, USA)



ORGANIZERS (UDS)

Heiko Rieger
Ludger Santen
Manfred Lücke

SCIENTIFIC ADVISORY BOARD (UDS)

Ivan Bogeski
Volkhard Helms
Markus Hoth
Karsten Kruse
Franziska Lautenschläger
Albrecht Ott
Heiko Rieger
Ludger Santen
Ralf Seemann

http://www.cell-physics.uni-saarland.de

SFB 1027 DFG GRK 1276 UNIVERSITÄT DES SAARLANDES

Pre-Conference Program

Monday, 23.9.		
09:00-09:30	Alexander Dreher	Spiral actin-polymerization waves can generate amoeboidal cell crawling
09:30-10:00	Hendrik Hähl	Hydrophobins at interfaces: Model proteins for the study of protein biofilms
10:00-10:30	Henrik Peisker	The extracellular adherence protein *of Staph. aureus* - new functions of a multifunctional adhesin
10:30-11:00		COFFEE
11:00-11:30	Jean-Baptiste Fleury	New Strategy to Study SNARE Mediated Membrane Fusion
11:30-12:00	Manuel Worst	*In vitro* Transcription and Translation: Epigenetic Regulation and Incorporation of non-canonical Amino Acids
12:00-12:30	Jörn Walter	Processes influencing the (in)stable copying of DNA-methylation patterns in mammalian cells
12:30-14:00		LUNCH
14:00-14:40	Guntram Bauer	Opportunities for international collaboration in basic research
14:40-15:10	Ivan Bogeski	Calcium and Reactive Oxygen Species as determinants of immune and skin cell function
15.10-15.40	Antonio Yarzagaray	v-SNARE-based protein-lipid interactions in Ca^{2+}-triggered exocytosis
15:40-16:00		COFFEE
16:00-16:40	Franziska Lautenschläger	Vimentin in amoeboid cell migration
16:40-17:20	Niels de Jonge	Studying protein complexes on whole cells in liquid using scanning transmission electron microscopy
17:20-18:00		MEMBER'S MEETING

Conference Program

Tuesday, 23.9.			
08:50-09:00		WELCOME	
09:00-09:45	Jacques Prost	Cell cortex and cell division	I.18
09:45-10:30	Richard McKenney	BicD2 and dynactin convert a non-processive cytoplasmic dynein to an ultra-processive directional motor	I.13
10:30-11:00		COFFEE	
11:00-11:30	Jian Liu	Traction oscillation confers focal adhesion mechanosensing – A tale of two distinct actin networks	C.11
11:30-12:00	Daniel Riveline	A simple active gel *in vivo* : the cytokinetic ring	C.14
12:00-12:30	Karsten Kruse	Length-distribution of actin filaments in the submembranous cortex of living cells	C.10
12:30-14:00		LUNCH	
14:00-14:45	Steven Gross	Control of motor activity: a new activation pathway	I.4
14:45-15:30	Reinhard Lipowsky	Multiscale motility of biomolecular machines	I.12
15:30-16:00		COFFEE	
16:00-16:45	Richard S. Lewis	Self-organization of store-operated calcium channels at the immune synapse	I.11
16:45-17:30	David I. Yule	A multi-scale modeling approach to better understand salivary gland fluid secretion	I.20
17:30-18:00	Markus Hoth	Combining experimental data and theoretical models to analyze cytotoxicity of primary human killer cells	C.6
18:00-21:00		POSTER SESSION	

Wednesday, 24.9.			
09:00-09:45	Josef A. Käs	Are biomechanical changes necessary for tumor progression?	I.10
09:45-10:30	Nicolas Minc	Developmental morphogenesis of a single cell	I.14
10:30-11:00		COFFEE	
11:00-11:30	Jaume Casademunt	Formation of helical membrane tubes around microtubules by monomeric kinesin	C.2
11:30-12:00	Falko Ziebert	The mystery of circling microtubules: tubulin lattice switches under force	C.15
12:00-12:30	Jan Kierfeld	Feedback mechanism for microtubule length regulation by bistable stathmin gradients	C.7
12:30-14:00		LUNCH	
14:00-14:45	Daniel J. Needleman	Self-Focusing of the Ran Gradient in mitosis: signaling, mechanics, and spindle size	I.15
14:45-15:30	Frank Jülicher	Phase separation in the cell cytoplasm	I.9
15:30-16:00		COFFEE	
16:00-16:45	Vincent Hakim	Cell motion in confined environments: collective modes and fluctuations	I.5
16:45-17:30	Roland Wedlich-Söldner	A pulsatile acto-myosin network organizes the apical surface of epithelial cells	I.19
17:30-18:00	Cécile Appert-Rolland	Microtubule based bidirectional transport	C.1
18:30-…		BARBECUE	

Thursday, 25.9.			
09:00-09:45	Paul Janmey	Non-linear elasticity and relaxation in polymer networks and soft tissues	I.7
09:45-10:30	Lars Hufnagel	Bioimaging embryonic development with light-sheet microscopy	I.6
10:30-11:00		COFFEE	
11:00-11:30	Thomas Risler	Tension-oriented cell divisions in zebrafish epiboly	C.13
11:30-12:00	Christoph Erlenkämper	Mechanics of cell rounding and apical migration in epithelial tissue	C.5
12:00-12:30	Stefan Klumpp	Motility and magnetism in magnetotactic bacteria	C.8
12:30-14:00		LUNCH	
14:00-14:45	Ian Parker	Imaging calcium signaling in intact cells down to the single-molecule scale	I.17
14:45-15:30	Sonia Cortassa	Insights from a computational-experimental synergy to elucidate disease mechanisms	I.3
15:30-16:00		COFFEE	
16:00-16:45	Roy Bar-Ziv	Programmable on-chip DNA compartments as artificial cells	I.1
16:45-17:30	José N. Onuchic	Decision making at the cell level: from microorganisms to possibly cancer	I.16
17:30-18:00	Jens Elgeti	Simulating growing tissues	C.4
19:00-…		SPEAKER DINNER	

Friday, 26.9.			
09:00-09:45	Françoise Brochard	Collective migration of multicellular aggregates	I.2
09:45-10:30	Jean-Francois Joanny	Physics of epithelial cell layers	I.8
10:30-11:00		COFFEE	
11:00-11:30	Hans-Günther Döbereiner	Foraging via topological phase transitions in the slime mold physarum polycephalum	C.3
11:30-12:00	Zdeněk Petrášek	Reaction-diffusion models of bacterial *min* protein dynamics	C.12
12:00-12:30	Moritz Kreysing	Thermal dis-equilibrium as a metabolic platform to allow for the evolution of life	C.9
12:30-14:00		CLOSING/LUNCH	

Poster List

P 1	Ackermann, Björn	ssDNA hybridisation in competition
P 2	Alansary, Dalia	Inhibition of SOCE by ROS is mediated by direct oxidation and altered kinetics of Orai1 protein diffusion
P 3	Astania, Ksenia	Intercellular communication between endothelial cells via tunneling nanotubes
P 4	Alert, Ricard	Physics of membrane-cortex adhesion and bleb nucleation
P 5	Aouane, Othmane	Boundary integral formulation for Stokes flow: application to the flow of red blood cells
P 6	Bandmann, Vera	Membrane processes in biomineralization
P 7	Barghash, Ahmad	Intragenic epigenetic modifications modulate exon usage in human across developmental stages
P 8	Baumgart, Johannes	The Xenopus spindle as an active liquid crystal
P 9	Becker, Björn	KDEL receptor dynamics, internalization and retrograde transport in mammalian and yeast cells
P 10	Belkacemi, Thabet	TRPC channels mediate Ca^{2+} influx and migration of astrocytes
P 11	Bleymehl, Katherin	Chemosensory responses of Grueneberg Ganglion Neurons depend on CNGA3
P 12	Bogeski, Ivan	The role of ORAI channels and their interplay with NADPH oxidase 2 in bacterial peptide-induced innate immune responses
P 13	Bonny, Mike	Formation of long lived local protein kinase C clusters after short Calcium puffs
P 14	Clavería, Viviana	Clustering of red blood cells in microcapillaries under flow conditions

P 15	Colanesi, Sarah	Managing oral biofilm formation with plant extracts
P 16	Dasgupta, Sabyasachi	A mechanistic understanding towards cellular uptake
P 17	de Jonge, Niels	Scanning transmission electron microscopy of whole eukaryotic cells in liquid
P 18	Dembla, Ekta	Biogenesis of large dense core vesicles in the adrenal chromaffin cells of newborn mice
P 19	Dhara, Madhurima	v-SNARE-based protein-lipid interactions in exocytosis
P 20	Dreher, Alexander	Spiral actin-polymerization waves can generate amoeboidal cell crawling
P 21	Eckrich, Tobias	Spontaneous Ca^{2+} signals in the mouse inner ear during development
P 22	Fleury, Jean-Baptiste	Interaction of bilayers in microfluidics: shedding light on the fast formation of single hemifused state
P 23	Fleury, Jean-Baptiste	How to Study a Single SNARE Mediated Membrane Fusion Event ?
P 24	Flormann, Daniel	Aggregation of red blood cells
P 25	Gadelha, Hermes	The flagellar counterbend phenomenon
P 26	Gamayun, Igor	Mechanism of Ca^{2+} leak from endoplasmic reticulum
P 27	Gartmann, Julia	Regulation of the cation channel protein Yvc1 by SUMOylation
P 28	Giehr, Pascal	Decoding of processes influencing the (in)stable copying of DNA methylation patterns in mammalian cells
P 29	Hafner, Anne	Intermittent motion of motor proteins in biological environments
P 30	Hähl, Hendrik	Protein films at tailored interfaces: Towards controlling adsorption and arrangement

P 31	Hamed, Mohamed	Integration of genetic and epigenetic data into cellular networks identifies major determinants associated with breast invasive carcinoma
P 32	Hoth, Markus	The calcium dependence of target cell killing by primary human immune cells
P 33	Hudalla, Philipp	Numerical analysis of hydrophobin adsorption at interfaces
P 34	Inamdar, Mandar	Mechanical modeling of collective sheet migration
P 35	Johann, Denis	Dynamics of diffusing particles interacting with directionally moving particles on a polar filament
P 36	Jung, Philipp	*Staphylococcus aureus*: cell wall elasticity and turgor pressure
P 37	Kaiser, Elisabeth	Conditional and selective activation of the Gq pathway in the heart and its impact on cardiac function in vivo
P 38	Karsch, Susanne	Mechanics of the nucleus dictated by the cytoskeleton
P 39	Klein, Sarah	Environmental control of stochastic cargo transport by teams of molecular motors.
P 40	Knörck, Arne	Ca^{2+} dependence of $CD8^{+}$ T cell proliferation and subset development
P 41	Konrad, Maik	Calcium and ROS: from detection to function
P 42	Kummerow, Carsten	A technique to distinguish two modes of immune cell killing on single cell level
P 43	Kummerow, Carsten	Quantitative analysis of calcium dependent migration in human killer cells
P 44	Kyriakopoulos Charalampos	Markov models for the (in)stable copying of DNA-methylation patterns in mammalian cells
P 45	Lannoo, Bruno	Generation of oscillating gene regulatory network modules
P 46	Lee, Po-Hsien	Brownian dynamics simulations of the molecular encounter at the *E. coli pap* promoter

P 47	Löhfelm, Aline	Visualising immediate effects of neurosteroids DHEA and 7β-OH-DHEA on neurons by calcium imaging
P 48	Lück, Alexander	Exact and numerical results for a simple microtubule length regulation model
P 49	Miederer, Anna-Maria	A novel STIM2 splice variant functions as a break for STIM mediated activation of Orai calcium channels
P 50	Mohammadi, Mina	DNA hybridization and kinetics of hairpin-loop molecules
P 51	Nazarieh, Maryam	Identification of Gene Regulatory Networks Governing Cellular Identity
P 52	Neef, Marc	Roll instabilities of an active polar fluid in the planar Taylor-Couette geometry
P 53	Nirody, Jasmis	ATP concentration regulates cellular processes in time and space
P 54	Nitze, Katja	Properties of novel voltage sensitive dyes in adult ventricular myocytes
P 55	Oriola, David	Subharmonic oscillations of collective molecular motors
P 56	Paknikar, Aishwarya	Dynamics of human blood platelets on elastic and patterned substrates
P 57	Peckys, Diana	Detecting protein complex subunits in whole eukaryotic cells in aqueous environment
P 58	Peglow, Martin	Interplay of channels, pumps and organelle location in calcium microdomain formation
P 59	Peinelt, Christine	The role of Orai3 in SOCE of prostate cancer cells
P 60	Peisker, Henrik	The extracellular adherence protein of *Staphylococcus aureus* - new functions of a multifunctional adhesin
P 61	Pick, Tillmann	Pharmacological regulation of the Ca^{2+} homeostasis in the endoplasmic reticulum
P 62	Rüdiger, Sten	Fast and slow excitability in models of intracellular calcium release

P 63	Sabass, Benedikt	Cooperative motion of bacteria on soft surfaces
P 64	Sain, Anirban	Shape transformation during endospore formation in bacteria.
P 65	Sander, Mathias	Living cell rheology: Fourier modes and spontaneous dynamic symmetry breaking
P 66	Schenkelberger, Marc	Competition for binding leads to ultrahigh oligonucleotide molecular recognition specificity
P 67	Scherer, Sabrina	Spontaneous autocatalysis in a primordial broth
P 68	Schorr, Stefan	The passive Ca^{2+}-efflux from the endoplasmic reticulum through the Sec61 complex: Regulation and physiological consequences
P 69	Schröder, Laura	Less-invasive transverse aortic banding in mice for hypertrophy studies
P 70	Schwarz, Karsten	Investigation of intermittent search and transport strategies within spherical domains
P 71	Shaaban, Ahmed	Mechanistic Role of Q_{bc}-SNAREs in vesicular cargo release
P 72	Shaebani, Reza	Anomalous diffusion of self-propelled particles on directed random networks
P 73	Shaib, Ali	Molecular mechanisms of exocytosis of large dense core vesicles in dorsal root ganglion neurons
P 74	Soumya, S. S.	Probing the role of spatial anisotropy and heterogeneity in contractility and adhesion distribution during cell steering
P 75	Spengler, Christian	Influence of surface and subsurface properties on the structure and activity of adsorbed bactericidal proteins
P 76	Stankevicins, Luiza	The role of vimentin in cell migration under confinement
P 77	Suiwal, Shweta	Molecular and functional characterization of Munc119 in photoreceptor synapses
P 78	Tamiello, Chiara	A microfabricated modular strategy for probing the impact of physical cues on adhesive cells

P 79	Tanimoto, Hirokazu	Aster migration in Sea Urchin eggs studied by 3D tracking and cell shape manipulation
P 80	Tarle, Victoria	Modelling the instability of an expanding cellular mono-layer during collective cell motion
P 81	Thewes, Nicolas	Hydrophobic interaction governs unspecific adhesion of staphylococci: a single cell force spectroscopy study
P 82	Tinschert, René	Ca^{2+} leak channels in the endoplasmic reticulum (ER)
P 83	Turlier, Hervé	Furrow constriction in animal cell cytokinesis
P 84	Vandanapu, Rama Ramesh	Heterologous expression and characterization of N-terminal domains of NOX5 : a calcium dependent NADPH oxidase
P 85	Visco, Paolo	Activity Driven Fluctuations in Living Cells
P 86	Vishavkarma, Renu	Role of actin filaments in correlating nuclear shape and cell spreading
P 87	Weber, Tobias	Motor-driven biological transport under confinement
P 88	Welter, Michael	Vascularization patterns and fluid flow in growing tumors
P 89	Wesseling, Mauro Carlos	Intracellular Ca^{2+} content and the regulation of phosphatidylserine exposure in human red blood cells
P 90	Wettmann, Lukas	Polar protein localization
P 91	Will, Thorsten	Identifying transcription factor complexes and their roles
P 92	Wollrab, Viktoria	Self-organisation of acto-myosin leads to closure of the cytokinetic ring in mammalian cells
P 93	Worst, Emanuel	Increasing complexity out of coexisting autocatalysts
P 94	Zhou, Xiao	Impact of bystander cells on cytotoxic efficiency of killer cells

P 95	Zimmer, Philipp	Evolution of increasingly complex linear molecules
P 96	Ziska, Anke	Novel strategies to characterize Sec61-associated proteins regulating the transport of precursor proteins into the mammalian ER

Abstracts of Invited Talks

I.1 Programmable on-chip DNA compartments as artificial cells

Roy Bar-Ziv

Department of Materials and Interfaces, Weizmann Institute of Science, Rehovot, Israel

We report assembly of silicon-fabricated, two-dimensional DNA compartments capable of metabolism, programmable protein synthesis, and communication. Metabolism is maintained by continuous diffusion of nutrients and products through a thin capillary, connecting protein synthesis in the DNA compartment with the environment. We programmed fundamental cellular functions including the self-regulation and periodic cycles of protein levels. Gene expression in the DNA compartment reveals a rich dynamical system that is highly controlled by geometry.

I.2 Collective migration of multicellular aggregates

Francoise Brochard, D. Cuvelier, S. Douezan, S. Dufour, J. Dumond, G. Beaune, D. Gonzales-Rodriguez, K. Guevorkian, V. Stirbat

Institut Curie, Paris, France

Single cell migration has been studied extensively but processes such as embryonic development or tumors often require the motion of a group of cells, which are mechanically coupled through cell-cell

contacts as well as through their substrate. We study here the collective motility of thousands of cells on soft gels decorated with fibronectin, whose motion can be 1) induced by a chemical or a rigidity gradient or 2) Spontaneous "running" aggregates The spreading dynamics of the monolayer expanding outwards from the aggregates is optimal in a narrow range of rigidity, where the diffusion coefficient presents a maximum. In this range of rigidity, we observe a symmetry breaking of cells polarity causing the aggregate to move spontaneously.

We observe by Confocal Optical Microscopy both the motion of the cells, and the displacement of the fluorescent beads encapsulated in the gel. Using the PIV method (particle image velocimetry) and the FFTC method (Fourier transform traction cytometry), we obtain the velocity and the force field maps of the moving aggregates.

I.3 Insights from a computational-experimental synergy to elucidate disease mechanisms

Sonia Cortassa

Center for Computational Medicine, Johns Hopkins School of Medicine, Baltimore MD, USA

In this presentation I will discuss two examples to illustrate the synergy between computational and experimental works to unravel mechanisms relevant for human disease.
The first example deals with the mechanisms involved in synchronized mitochondrial oscillations in the heart that can lead to cardiac arrhythmias with fatal outcome. Novel mechanisms of mitochondrial coordination and function were unraveled with this combined

experimental-computational strategy. This work consisted in the successive and iterative reciprocal potentiation of the loops: simulation–validation and prediction–experimentation.
The second example involved the quantitative translation of high-throughput metabolomics into metabolic fluxes in the context of diabetes and heart function. Methodologically, the approach comprised an algorithmic platform consisting of several methods integrated to a computational model. As a bench test, the procedure devised was applied to understanding the mechanical performance of diabetic mice hearts perfused with high glucose, fatty acids and β-adrenergic stimulation. The results obtained will be discussed in the context of the positive acute effects of fatty acids on redox balance and cardiac contractility in diabetic mice.
The two examples presented underscore the important role played by computational modeling in the quantitative interpretation of experimental data leading to a deeper mechanistic understanding of complex biological phenomena.

I.4 Control of motor activity: a new activation pathway

Steven Gross

University of California, Irvine CA, USA

Past work on regulation of kinesin had predominantly focused on control of motor activity via modulation of the head-tail interaction, which is believed to be predominantly relieved upon recruitment of the motors to their cargoes. We have discovered a new pathway, regulating kinesin function, independent of the head-tail interaction. I will provide an overview of this new system, and discuss open question as well.

I.5 Cell motion in confined environments: collective modes and fluctuations.

Vincent Hakim

Département de Physique, École Normale Supérieure, Paris, France

In different biological processes, cells move in a coordinated way. Several experiments have quantitatively investigated this phenomenon. We will describe a simple model of interacting persistent random walkers that has been developed to phenomenologically describe collective cell motion, based on data obtained by the team of P. Silberzan (Institut Curie, Paris). The model helps to explain the observed motion of confined cell assemblies, which displays stochastic reversals of global rotational motion and pulsatile collective modes.

I.6 Bioimaging embryonic development with light-sheet microscopy

Lars Hufnagel

European Molecular Biology Laboratory, Heidelberg, Germany

Developmental processes are highly dynamic and span many temporal and spatial scales. A whole-embryo view of morphogenesis with subcellular resolution is essential to unravel the interconnected dynamics at the varying scales of development, from interactions within cells to those acting across the whole embryo. Bridging scales from the submicron to the millimeter range with a temporal resolution of several seconds (combined with a total imaging time of several hours) not only poses tremendous challenges for modern microscopy methods but also requires powerful computational approaches for data handling, processing and image analysis. I present a multiview selective-plane illumination microscope (MuVi-SPIM), comprising two detection and illumination objective lenses, that allows rapid /in toto/ fluorescence imaging of biological specimens with subcellular

resolution. The fixed geometrical arrangement of the imaging branches enables multiview data fusion in real time. The high speed of MuVi-SPIM allows faithful tracking of nuclei and cell shape changes, which we demonstrate through /in toto/ imaging of the embryonic development of /Drosophila melanogaster/.

I.7 Non-linear elasticity and relaxation in polymer networks and soft tissues

Paul Janmey

Institute for Medicine and Engineering, University of Pennsylvania PA, USA

The stiffness of tissues in which cells are embedded has effects on cell structure and function that can act independently of or override chemical stimuli. Most measurements of tissue stiffness report elastic moduli measured at a single frequency and at a low strain, but such measurements are inadequate to define the mechanical environment to which cells respond in vivo. Tissues and the cells within them are subjected to a range of strains in vivo depending on their elastic modulus and the forces applied to them. These strains often exceed the range of linear viscoelasticity that is reflected in measurements at small strains. Rheologic measurements of liver, brain, and adipose tissues over a range of shear, compressive, and elongational strains show that the viscoelastic response of these tissues differs from that of synthetic hydrogels or blood clots that have similar elastic moduli when measured in the linear range. The shear moduli of soft tissues generally decrease with increasing shear or elongational strain, but they strongly increase under uniaxial compression. In contrast, networks of crosslinked collagen or fibrin soften under compression, but strongly increase shear modulus when deformed in extension. The mechanisms leading to the unusual strain-dependent rheology of soft tissues and fibrous networks do not appear to be explained by current models of polymer mechanics, but appear to relate to local and global volume conservation within the networks and tissues.

I.8 Physics of epithelial cell layers

Jean-Francois Joanny

Institut Curie, Paris, France

We discuss some physical properties of epithelial cell layers focussing on the intestinal epithelium. We first present a 3 dimensional vertex model to predict cell shape in the epithelium. The structure of the epithelium is found by minimisation of an effective energy which includes the various interfacial energies of a cell with its environment. There are 3 possible cell shapes: columnar (elongated in the direction perpendicular to the epithelium), squamous (spread along the epithelium) or cuboidal. We discuss the transitions between these shapes that can be continuous and go through a cuboidal shape or discontinuous and allow in certain range of parameters for co-existence between columnar and squamous cells. We then discuss bent epithelial layer and the formation of hollow cell tubes and hollow cell spheres (cysts). Finally, we consider the role of stem cells and cell differentiation in the epithelial layers and the formation of niches for the various types of cells.

I.9 Phase separation in the cell cytoplasm

Frank Jülicher

Max Planck Institute for the Physics of Complex Systems, Dresden, Germany

Cells exhibit a complex spatial organization, often involving organelles that are surrounded by a membrane. However, there exist many structures that are not membrane bounded. Examples are the centrosome, meiotic and mitotoc spindles as well as germ granules. An interesting question is how such structures are assembled in space inside the cytoplasm which is essentially a fluid where all components usually mix? I will highlight the importance of phase coexistence and phase separation of fluid phases in the cell cytoplasm as a basic

mechanism of the spatial organization of cells. Droplets in the cytoplasm can represent microreactors with different composition and chemistry as compared to the surrounding cytosol. I will discuss how such concepts shed light on the structure and organization of centrosomes and of germ granules.

I.10 Are biomechanical changes necessary for tumor progression?

Josef A. Käs

Institute for Experimental Physics I, University of Leipzig, Leipzig, Germany

Already the Roman Celsus recognized rigid tissue as characteristic for solid tumors. Conversely, changes towards a weaker cytoskeleton have been described as a feature of cancer cells since the early days of tumor biology. It remains unclear if a carcinoma's rigid signature stems from more inflexible cells or is caused by the stroma. Despite that the importance of cell biomechanics for tumor progression becomes more and more evident the chicken-and-egg problem to what extent cancer cells already change their mechanical properties within the solid tumor in order to transgress its boundary or mechanical changes are induced by the microenvironment when the cell has left the tumor has been discussed highly controversial. Comprehensive clinical biomechanical measurements only exist from tumor tissue without the possibility to identify individual cells or from individual cancer cells from pleural effusions. Since the biomechanical properties of cells in carcinomas remain unknown measurements on individual cells that directly stem out of primary tumor samples are required, which we have conducted. We found in cervix and mammary carcinomas a distinctive increase of softer cells as well as contractile cells. A soft and contractile cell is like a strong elastic rope. The cell can generate a strong tensile tension to pull its self along and is soft against compression to avoid jamming.

I.11 Self-organization of store-operated calciumchannels at the immune synapse

Richard S. Lewis

Department of Molecular and Cellular Physiology, Stanford University School of Medicine, Stanford CA, USA

When T lymphocytes encounter and recognize antigens on the surface of antigen-presenting cells (APCs), they spread out on the APC surface to form a highly organized intimate connection called an immune synapse. Dynamic rearrangements of the actin cytoskeleton at the synapse are critical for the T cell antigen receptor to activate phospholipase C, generate IP3 and open store-operated calcium (CRAC) channels, leading ultimately to T cell activation. The ER Ca^{2+} sensor STIM1 and the CRAC channel protein Orai1 localize to the synapse, where they self-organize in two ways. At sites where the ER comes within 10-20 nm of the plasma membrane (PM) to form ER-PM junctions, STIM1 and Orai1 accumulate through a diffusion-trap mechanism driven by the formation of STIM-Orai complexes. Our recent studies using single-molecule tracking validate this model and reveal an unexpected degree of protein dynamics within junctions and across junctional boundaries. The second mode of self-organization arises from crosstalk between CRAC channels and the actomyosin cytoskeleton. Using an in vitro model of synapse formation, we observe that microtubules continually project ER tubules towards the periphery of the synapse, but that radial retrograde flow of actomyosin sweeps the ER tubules and STIM1 and Orai1 clusters back towards the center, such that STIM1-Orai1 clusters ultimately accumulate in a central actin-poor region. Interestingly, Ca^{2+} entry through open CRAC channels acts to sustain this retrograde actomyosin flow, thus creating a Ca^{2+}-dependent feedback loop that self-organizes active CRAC channel complexes and the ER at the center of the immune synapse. Underlying mechanisms and functional consequences of this crosstalk for the regulation of T cell activation will be discussed.

I.12 Multiscale motility of biomolecular machines

Reinhard Lipowsky

Max Planck Institute of Colloids and Interfaces, Potsdam-Golm Science Park, Golm, Germany

The living cell contains a large variety of biomolecular machines that use chemical free energy for intracellular transport and remodelling as well as for information processing. A bottom-up approach from simple to more complex systems will be used and applied to the following processes; free energy transduction of single cytoskeletelal motors, cooperative transport by teams of motors, dynamics of actin filaments, and protein synthesis by ribosomes.

I.13 BicD2 and dynactin convert a non-processive cytoplasmic dynein to an ultra-processive directional motor

Richard J. McKenney, Walter Huynh, Marvin E. Tanenbaum, Gira Bhabha, and Ronald D. Vale.

Department of Cellular and Molecular Pharmacy, UCSF School of Medicine, San Francisco CA, USA

Cytoplasmic dynein is the predominant minus-end directed microtubule motor in metazoan cells. Dynein transports diverse cargoes over long distances in neurons, and the motor is thought to be adapted for a myriad of cellular functions through the use of several accessory protein factors that impinge on its basic biophysical characteristics. One of these accessory factors is the multisubunit dynactin complex, which has been implicated in dynein-based cargo transport and the modulation of dynein processivity and directionality. While isolated dynein from Saccharomyces has been shown to be a strongly processive motor, dynein from other organisms displays weakly processive, bidirectional or diffusive motility. Here we show

that, on its own, cytoplasmic dynein from humans and other metazoans is not a processive motor. Previous attempts to study dynein-dynactin co-complexes have found relatively modest effects on dynein processivity and directionality by dynactin. We utilize the evolutionarily conserved coiled-coil adapter protein BicD2 to strongly induce the formation of a stable dynein-dynactin-BicD2 (DDB) supercomplex that is over 2MDa in size. Using multicolor single-molecule microscopy, we have found that, remarkably, the purified DDB supercomplex is unidirectional and ultra-processive, displaying run-lengths that greatly exceed the previously observed enhancement of single dynein run-lengths by dynactin. Our data indicate a role for the dynactin microtubule-binding domain in ultra-processive motility. Furthermore, we have found that BicD2 is one of a number of coiled-coil adapter proteins that link dynein to dynactin and activate processive motility. Mutations in all three of the DDB components cause human neurodegenerative diseases, highlighting the critical role of processive retrograde motility in human health. Our data suggest that the dynein motor is more plastic that previously thought, able to transition from a non-processive to an ultra-processive mode of motility upon association with external regulatory factors. Further, our data indicate that activation of dynein is coupled to cargo selection.

I.14 Developmental morphogenesis of a single cell

Nicolas Minc

Institut Jacques Monod, Université Paris Diderot/CNRS, Paris, France

How cells establish their proper morphogenesis is a fundamental biophysical problem. I will present recent work investigating the dynamic development of cellular form in the rod-shape fission yeast cell. These studies are based on monitoring how small symmetric fission yeast spores grow and self-organize to break symmetry for the definition of their very first polarity axis. In a first part, I will highlight interplays between surface mechanics of the spore cell wall and the stability of Cdc42-based polarity caps which control spatio-temporal aspects of spore symmetry breaking. In a second part, I will discuss mechanisms by which these initial polarity caps control their width and

adapt it to cell surface curvature, a process likely relevant to understand how functional cortical domains scale to cell size.

I.15 Self-focusing of the Ran Gradient in mitosis: signaling, mechanics, and spindle size

Daniel J. Needleman

Harvard SEAS, Cambridge MA, USA

During spindle assembly, microtubules are highly enriched near chromatin by a process which, in many systems, is driven by the GTPase Ran. The Ran pathway has been proposed to establish a reaction-diffusion network that generates gradients in the behaviors of soluble proteins around chromatin, but the manner in which this happens is poorly understood. To better characterize the behavior of the Ran pathway, we developed a novel form of fluorescence fluctuation spectroscopy capable of quantitatively measuring the concentration, diffusion, and interactions of soluble proteins simultaneously at hundreds of locations throughout cells. We use this technique to study the behaviors of soluble Ran, importin-alpha, importin-beta, RanBP1, RanGAP, and a variety of downstream cargo proteins throughout mitotic human tissue culture cells, and we investigate how the spatial organization of this network changes in response to perturbations. Our results suggest that a self-focusing of the Ran pathway is produced by an interplay between soluble gradients of upstream signaling molecules and the mechanics of the microtubule network they generate. This feedback has interesting implications for models of spindle assembly and the maintenance of spindle size.

I.16 Decision making at the cell level: from microorganisms to possibly cancer*

José N. Onuchic

Center for Theoretical Biological Physics, Rice University, La Jolla, USA

In nature, bacteria are living in large colonies whose number of bacteria may reach up to 100 times the number of people on Earth. The new research showed how under life alarming conditions the bacteria in the colony communicate via chemical messages and how each bacterium performs a sophisticated decision process by using a specialized network of genes and proteins. This complex network enables to perform complex calculations to assess the prose and cones of the different choices using guided by a new principles of game theory.

Many bacteria respond to extreme stresses, such as starvation, poisons, damage to DNA, etc. by creating spores - dormant versions that are highly resistant. The spores wait and germinate if/when the stress is removed. This process involved more than 500 genes, and takes about 10 hours in the studied bacteria - Bacillus subtilis. Sporulation ends with the death of the mother cell after a copy of the DNA is stored in a special capsule – the spore. The mother cell then break open and its DNA and left over proteins are released to the environment. The bacteria on the road towards sporulation have the option to decide to change their fate and escape into a different state called competence.

This is where game theory enters. Bacteria game theory if far more advanced than the well-known case of the Prisoner Dilemma. Classic prisoner's dilemma usually told for two prisoners given them an offer - if one does not admit the crime and second, it will be admitted two years' imprisonment and not admitted will receive 6 years in prison. If they admit - they will get 4 years, and none of them admit, they were free. “Prisoner's dilemma” here is more complex," each bacterium must decide whether to become a spore (cooperate) or escape into competence (take advantage of the others) while it has a limited time to decide when a clock is ticking.

Connections between bacteria decision-making in a colony with cancer will be discussed. The idea that in both bacterial colonies and growing

tumors, genetic variability is selected by local conditions so as to increase growth will be explored.

*Supported by the National Science Foundation (NSF) and the Cancer Prevention and Research Institute of Texas (CPRIT)

I.17 Imaging calcium signaling in intact cells down to the single-molecule scale

Ian Parker

Departments of Neurobiology & Behavior, Physiology & Biophysics, University of California, Irvin CA, USA

Calcium serves as a universal intracellular messenger, controlling cellular processes as diverse as gene transcription, secretion, and electrical excitability. This versatility arises through the mechanisms by which Ca^{2+} signals are generated and transmitted to act over very different time and distance scales, ranging from waves with periods of minutes to transient domains at nanometer and millisecond scales. We focus on Ca^{2+} signals generated by clusters of inositol triphosphate (IP3) receptor/channels that release Ca^{2+} from the endoplasmic reticulum into the cytosol. Channel opening is regulated by Ca^{2+} itself, creating positive and negative feedback loops that result in a hierarchy of signals ranging from openings of single channels and concerted openings of channels in a cluster to waves that sweep throughout a cell. The spatiotemporal patterns of cellular Ca^{2+} signals thus depend on the properties of the IP3 receptors, their spatial arrangement in the cell, and their interactions via Ca^{2+} diffusion and other mechanisms. We are studying these mechanisms utilizing novel optical imaging techniques to resolve the functioning of individual IP3 receptor/channels in intact cells, and to localize and track single IP3 receptor proteins with nanometer precision.

I.18 Cell cortex and cell division

Jacques Prost

ESPCI and Curie Institute, Paris, France

Much of the cell mechanics, morphology and motility is determined by the dynamical properties of an actin network moving under the action of molecular motors and by a continuous process of polymerization/depolymerization called treadmilling. The actin network constitutes a physical gel the cross-links of which are both temporary and mobile. It is more complex than a physical gel in that it has a macroscopic polarity due to the microscopic polarity of actin filaments and in that the cross-links are dynamically redistributed by molecular motors. Most of this network is confined in a thin region below the plasma membrane and called “actin cortex”. I will explain how one can propose equations describing this situation and develop a useful analogy with wetting phenomena in physics. In conventional wetting the proximity of a phase transition plays an important role and is controlled by temperature or pressure, here the control is contractility. I will then consider how cortices facing at a distance can interact and give a few examples concerning cell dynamics such as cell division.

I.19 A pulsatile acto-myosin network organizes the apical surface of epithelial cells

Christoph Klingner, Anoop V. Cherian, Johannes Fels, Roland Wedlich-Söldner

Institute of Cell Dynamics and Imaging, University of Münster, Münster, Germany

Although it is widely accepted that the cortical actin cytoskeleton plays a role in cell mechanics and cellular morphogenesis, there is surprisingly little information on cortex organization at the apical

surface of mammalian cells. We have recently characterized the spatial organization and dynamics of a previously unappreciated acto-myosin network at the apical surface of Madin-Darby canine kidney (MDCK) cells. In contrast to the well described dense array of short and static microvilli (MV) in confluent MDCK cells, the apical surfaces of non-confluent MDCK cells form highly dynamic protrusions, which exhibit complex and spatially correlated patterns of reorganization that are dependent on myosin II activity. Surprisingly, myosin II itself is organized into an extensive network of filaments spanning the entire apical membrane in non-confluent epithelial cells. Dynamic MV and apical myosin filaments form an interconnected, pre-stressed network and exhibit highly coordinated motility. Interestingly, this network both restricts and facilitates lateral mobility of apical membrane probes such as collagen-coated beads and the EGF receptor, suggesting that coordinated acto-myosin dynamics provides the basis for apical cell membrane organization.

I.20 A multi-scale modeling approach to better understand salivary gland fluid secretion

David I. Yule

Department of Pharmacology and Physiology, University of Rochester Medical School, Rochester NY, USA

The secretion of saliva provides hydration and lubrication to the oral cavity while the protein content contributes enzymes, which begin to digest food, and additional proteins, which protect the oral cavity and upper gastrointestinal tract from bacterial and fungal infections. Disease such as Sjogrens Syndrome or following γ-irradiation therapy for head and neck cancer result in profound xerostomia or “dry-mouth”. These patients are subject to rampant oral infections, have difficulty speaking and assimilating food, which results in a significant loss of their quality of life. The rate-limiting step for fluid secretion is the vectoral secretion of NaCl across the apical plasma membrane of acinar cells stimulated by a rise in intracellular Ca^{2+}. This “primary” fluid is then modified by ion absorption and secretion by the ductal

epithelium. We have used a combination of experimental investigation and quantitative theoretical modeling to further our understanding of both salivary gland physiology and pathology in a manner that neither single approach can easily accomplish alone. To this end, a multi-scale model of fluid secretion is being developed from experimental data, which captures the essential spatial and temporal mechanistic features required for fluid flow at the molecular, cellular and glandular levels. The approach uses a process of iterative testing between model predictions and experimentally determined parameters and outcomes. The power of this approach is that the model is not only used to quantitatively explain and interpret the experimentally derived data, but perhaps more importantly, also can suggest further experiments and subsequently be used to predict their outcomes. In the presentation, examples will be used to illustrate the key elements of the model development, together with demonstrating its' predictive ability. It is envisioned that the model will continue to further our understanding of physiology of fluid secretion and by adaptation with parameters obtained under conditions of dry mouth may ultimately suggest novel therapies to restore salivary gland function that would not be readily evident from traditional, purely experimental methodologies.

Abstracts of Contributed Talks

C.1 Microtubule based bidirectional transport

Cécile Appert-Rolland

Laboratoire de Physique Théorique, CNRS, Université Paris-Sud, Orsay, France

It is not clear yet how microtubule based transport can be organized in an efficient way, given the crowded environment and the presence of cargos going in opposite directions while using the same tracks. Here we propose two approaches to contribute to this question.
First, we focus on one single cargo-motors system. We model it by taking explicitely into account the position of the various motors on the track [1] and explore the bidirectional cargo dynamics that result from the tug-of-war between oppositely going motors. We show that the drift of cargo can be controlled, and even reversed, by external parameters such as the viscous force exerted on the cargo [2]. In the cell, obstacles could play the role of such an effective viscosity. We find superdiffusion on short timescales, indicating some motor cooperation induced by cargo-mediated coupling. We will discuss all our results in view of experimental observations. The condition of existence of high motility states will also be explored.
In the cell, many such cargos with opposite bias are moving on the same microtubule tracks. In a second stage, we will explore several scenarios that can contribute to the efficiency of collective motion, and that are based either on microtubules dynamics [3, 4], interactions between motors [5], or tuning of the bidirectionality of each cargo.

[1] S. Klein, C. Appert-Rolland, L. Santen: Stochastic modeling of cargo transport by teams of molecular motors. Proceedings of TGF'13 (Jülich 2013).

[2] S. Klein, C. Appert-Rolland, L. Santen. In preparation.

[3] M. Ebbinghaus, C. Appert-Rolland, L. Santen: Bidirectional transport on a dynamic lattice, Phys. Rev. E 82, (2010) p 040901.

[4] M. Ebbinghaus, C. Appert-Rolland, L. Santen: Bidirectional traffic on microtubules, Lecture Notes in Computer Science 6350 (2010) 542-551.

[5] M. Ebbinghaus, C. Appert-Rolland, L. Santen: Particle interactions and lattice dynamics: Scenarios for efficient bidirectional stochastic transport?, J. Stat. Mech. (2011) P07004.

C.2 Formation of helical membrane tubes around microtubules by monomeric kinesin

David Oriola[1], Sophie Roth[2,3], Marileen Dogterom[2,3], Jaume Casademunt[1]

[1]*Departament d'Estructura i Constituents de la Matèria, Universitat de Barcelona, Barcelona, Spain*

[2]*FOM Institute for Atomic and Molecular Physics (AMOLF), Amsterdam, The Netherlands*

[3]*Department of Bionanoscience, TU Delft, Delft, The Netherlands*

The kinesin-3 motor KIF1A is specific for the transport of synaptic vesicle precursors in axons, a very demanding task that involves carrying large cargoes along very long distances. In the densely crowded environment of axons, the capacity to collectively generate large forces may be crucial to overcome obstacles and possible traffic jams that may hinder the delivery of the cargo. Paradoxically, KIF1A motors are relatively inefficient and weak due to the presence of a weakly bound diffusive state in their processive cycle. In particular, its monomeric form is about 60 times weaker than conventional two-headed kinesin. In recent theoretical studies, it has been argued that, thanks to the diffusive state, these motors may be particularly adapted to cooperative action [D. Oriola & J. Casademunt, Phys. Rev. Lett. 111, 048103 (2013)]. By means of membrane tube extraction experiments, we show that monomeric KIF1A, despite its individual weakness, is indeed capable to collectively extract tubes, in conditions similar to those for experiments with conventional kinesin, thus proving their remarkable cooperativity. Moreover, our experiments have revealed an unexpected result, namely, that tubes extracted by KIF1A generically wind around the microtubules, showing that these motors collectively

exert significant transversal forces. We characterize experimentally the pitch of these helical tubes and their dynamics and propose a model to explain their basic features. Finally, we speculate on the possible relevance of transversal force generation to overcome roadblocks in axons, and to ensure long range collective processivity.

C.3 Foraging via topological phase transitions in the slime mold physarum polycephalum

HG Döbereiner, C. Oettmeier, A. Fessel, J. Lee, E. Bernitt

Institute for Biophysics, University Bremen, Bremen, Germany

In favorable environmental conditions, the slime mold *Physarum Polycephalum* percolates to form an extended transport network. We have recently described this percolation transitions using graph theory and found universal behavior for small link degree, PRL 109,078103 (2012). When food is sparse, percolation is suppressed in favor of the formation of independent globular satellites, which radially swarm out to explore the environment. We do find these novel satellites to be remarkable synchronized in their motile behavior. They progress in a keratocyte-like shape. The motile mechanism relies on an internal vein network, which is stationary with respect to the substrate. This vein network exhibits periodic shuttle streaming with a net transport of cytoplasm to the advancing front. After a view hours, satellites come to a halt and undergo a topological transition to a network via hole-formation in between veins.

C.4 Simulating growing tissues

Jens Elgeti

Institut of Complex Systems, Forschungszentrum Jülich, Jülich, Germany

Growth of solid tumors or metastasis requiers, besides massive biomedical changes, also a spatial remodeling of the tissue. This

remodeling, often including displacements of healthy tissue around, requires mechanical work to be done. These mechanics of growth has attracted a lot of attention in recent years, but still remains poorly understood.
We use particle based simulations to study mechanical properties and effects in growing and motile tissues. These simulations have been helpful in understanding, interpreting and designing experiments. I will present an overview of the simulation thechnique, and how it contributed to recent develobments in three dimensional tissue growth and collective cell migration. In a recent series of simulations and close experimental collaborations we found important interfacial and surface effects that lead to novel phenomena. For example, the tissue divides favorably at a free surface, even without any nutrient effects. This leads to the possibility and stability of a negative homeostatic pressure. In turn, a negative homeostatic pressure leads to naturally to finite steady states and tensile states.

[1] M.Basan et al, PNAS 110: 2452 (2013).

[2] F. Montel et al, N. J. Phys. 14: 055008 (2012).

[3] F. Montel et al, Phys. Rev. Lett. 107: 188102, (2011).

[4] M. Basan et al, Phys. Biol. 8: 026014, (2011).

[5] J. Ranft et al, PNAS 107: 20863, (2010).

C.5 Mechanics of cell rounding and apical migration in epithelial tissue

Christoph Erlenkämper, Maria Runjano, Renata Basto, and Jean-Francois Joanny

Institut Curie, Paris, France

Cell rounding and interkinetic nuclear migration prior to cell division are essential to maintain the structural integrity of pseudostratied epithelia such as the neuronal precursor cells in the developping brain of drosophila melanogaster larvae. We investigate the shape changes of dividing cells in groups of epithelial cells by stochastic and analytic means. We find that a homogeneous increase in surface tension of

dividing cells as compared to their neighbors leads to the rounding but not to the observed apical migration of cells. However, a gradient of actin contractility along the apico-basal axis qualitatively accounts for the *in vivo* conguration of the tissue before cytokinesis.

C.6 Combining experimental data and theoretical models to analyze cytotoxicity of primary human killer cells

Markus Hoth

Department of Biophysics, School of Medicine, Saarland University, Homburg, Germany

To mechanistically understand basic (physical) principles of primary human killer cell cytotoxicity against tumor cells is the major aim of our project in cooperation with the groups of Heiko Rieger and Karsten Kruse. The human immune system utilizes two cell types to kill tumor cells: cytotoxic T lymphocytes (CTL) and natural killer (NK) cells. Whereas tumor cell killing by CTL depends on the presentation of antigens through major histocompatibility complexes-I (MHC-I) on the surface of tumor cells, NK cells are specialized to kill tumor cells with low (= down-regulated) MHC-I expression. To escape immune surveillance is obviously a major goal of tumor cells and several mechanisms have evolved to optimize immune escape. MHC-I down-regulation is one such mechanism to escape CTL detection but at the cost of more efficient NK cell-based elimination.
To study single tumor cell killing, we have established novel assays to monitor in parallel apoptosis and necrosis of tumor cells by CTL and NK cells in 2- and 3-dimensional extracellular matrix-based environments. These assays are used for quantitative measurements of killer cell migration, killer-target cell contact formation, lytic granule release and tumor cell elimination. We are very interested to understand the efficiency of tumor cell killing and the basic mechanisms that determine this like for instance Ca^{2+} signaling, modulation of lytic granule release probability, search strategies or the role of innocent bystander cells for tumor cell killing. To achieve these

goals experimental data are included into theoretical models, from which predictions are derived to direct further experimental approaches and to facilitate the quantitative mechanistic understanding of tumor cell killing by CTL and NK cells with a special focus on efficiency.

C.7 Feedback mechanism for microtubule length regulation by bistable stathmin gradients

Jan Kierfeld and Maria Zeitz

Physics Department, TU Dortmund, Dortmund, Germany

Microtubule polymerization dynamics is regulated by the signaling proteins Rac1 and stathmin. In cells, the microtubule growth rate is inhibited by the soluble protein stathmin, which, in turn, is inactivated by Rac1. Growing microtubules activate Rac1 at the cell edge, which closes a positive feedback loop. Assuming that Rac1 is activated by microtubule contact at the cell edge, we formulate and analyze a model for microtubule growth regulated by Rac1 localized at the cell edge and cytosolic stathmin. For a homogeneous stathmin concentration in the absence of Rac1, we find a switch-like regulation of the microtubule mean length by stathmin. For constitutively active Rac1 at the cell edge, stathmin is deactivated locally, which establishes a spatial gradient of active stathmin. In this gradient, we find a stationary bimodal microtubule length distribution. One subpopulation of the bimodal length distribution can be identified with fast growing and long "pioneering" microtubules in the region near the cell edge, which have been observed experimentally. The biological feedback loop is closed through Rac1 activation by MTs. This establishes a bistable switch with two stable states: one stable state correponds to upregulated MT mean length and bimodal MT length distributions, i.e., "pioneering" MTs; the other stable state corresponds to an interrupted feedback with short MTs. Stochastic effects as well as external perturbations can trigger switching events.

C.8 Motility and magnetism in magnetotactic bacteria

Stefan Klumpp[1] and Damien Faivre[2]

[1]Department Theory & Bio-Systems, and [2]Department Biomaterials, Max Planck Institute of Colloids and Interfaces, Potsdam, Germany

Magnetotatic bacteria align along magnetic field lines with the help of magnetic organelles called magnetosomes. Magnetosomes contain magnetic nanoparticles (iron oxide or iron sulfide) and are aligned along a cytoskeletal structure, the magnetosome filament, to form a chain, which acts as a cellular compass needle. We introduce a model for the formation of the magnetosome chain and show that magnetic interactions between the nanoparticles are not sufficient for chain formation. Rather active transport towards the cell center is required in addition to magnetic attraction [1]. In addition, we use a combined theoretical and experimental approach to study how magnetic alignment helps the cells to swim in an oxygen concentration gradient towards the preferred micro-oxic zone, a behavior known as magneto-aerotaxis. The magnetic field can provide an axis and/or a direction for motility. A comparison of the magneto-aerotactic behavior of different strains of magnetotactic bacteria shows that some strains use the direction given by the magnetic field instead of sensing an oxygen concentration gradient and that such replacement can occur separately for low-oxygen and high-oxygen conditions [2].

[1] S. Klumpp and D. Faivre, PLoS One 7, e33562 (2012).

[2] C. Lefevre et al., submitted (2014).

C.9 Thermal dis-equilibrium as a metabolic platform to allow for the evolution of life

Moritz Kreysing[1,2], Lorenz Keil[1], Simon Lanzmich[1], Christof Mast[1], Stephan Krampf[1] and Dieter Braun[1]

[1]*Systems Biophysics, Ludwig Maximilian University, Munich, Germany*

[2]*Max Planck Institute for Molecular Cell Biology and Genetics, Dresden, Germany*

Life is accepted to have diversified as a result of Darwinian evolution. In order for the first genomic replicators to escape the destiny of equilibrium physics, decay, an early metabolism must have been in place to sustain the emergence of molecular complexity. To this end, we report on experimental findings that a mere temperature gradient across a sub-millimeter sized compartment can be used to filter bio-molecules from dilute solutions. Because the characteristics of this thermophoretically based filter are strongly non-linear with regard to polymer length, the through-flow system is able to accumulate exclusively long polymers, as firstly shown here by the length selective fractionation of solute DNA strands. Exploiting convectively driven temperature cycles [1], the locally accumulated DNA is additionally able to replicate in a PCR type manner. As we demonstrate, the combination of length selective accumulation and replication renders this compartment a niche in which heterogeneous populations of DNA strands are subject to a selection pressure in favor of molecular complexity, a so far unresolved prerequisite to the onset of Darwinian evolution [2].

[1] C. Mast and D. Braun, PRL, 104:188102 (2010).

[2] D. Mills, L. Peterson, S. Spiegelman, PNAS, 58:217 (1967).

C.10 Length-distribution of actin filaments in the submembranous cortex of living cells

M. Fritzsche[1], C. Erlenkämper[2], R. Thorogate[3], G. Charras[3], K. Kruse[2]

[1]*London Centre for Nanotechnology and Department of Physics and Astronomy, University College London, London, United Kingdom*

[2]*Theoretical Physics, Saarland University, Saarbrücken, Germany*

[3]*London Centre for Nanotechnology and Department of Cell & Developmental Biology, University College London, London, United Kingdom*

The submembranous cortex of animal cells is a polymeric network of actin filaments. The cortex is the main determinant of cell shape and mechanics, thus playing a vital role during cell division, cell migration, and tissue morphogenesis. Yet, little is known about its assembly kinetics and structural organization. In particular, knowledge of the cortical actin filament length-distribution is important for inferring cortical mechanics from polymer physics principles. We used single molecule imaging together with drugs perturbing actin turnover dynamics to quantitatively characterize actin filament assembly in the cortex of living cells. Employing the assembly rates determined in vivo, we performed a computational analysis of the essential turnover processes. From this, we deduced the presence of two filament subpopulations with distinct assembly mechanisms and average lengths differing by a factor 10. These findings open the way for understanding the dynamic interplay between actin turnover and cell mechanics.

C.11 Traction oscillation confers focal adhesion mechanosensing – A tale of two distinct actin networks

Zhanghan Wu, Sergey V. Plotnikov, Clare M. Waterman and Jian Liu

National Heart, Lung, and Blood Institute, NIH, Bethesda MD, USA

Focal adhesion (FA) is a dynamically formed structure, serving as the "foot" of migrating cells. Combining theory and experiment, we established the first coherent model of FA formation that integrates the coordinated action from branched actin network and stress fiber in FA mechanosensation. Upon interacting with the nascent FA, the retrograde actin flux gives rise to a traction peak near the FA distal tip, and enlarges the FA area that paves the way for FA-stress fiber engagement. The actomyosin contractility from stress fiber generates the second traction peak near the FA center. Due to the stress fiber elongation-mediated negative feedback with the actomyosin contractility, the central traction peak oscillates and optimizes the effective range over which FA traction force generation can precisely measure up the substrate stiffness. Our work thus sheds light on the mechanism of FA mechanosensing.

[1] Pelham, R.J. & Wang, Y.-l. Proc Natl Acad Sci 94, 13661-13665 (1997).

[2] Paszek, M.J. et al. Cancer Cell 8, 241-254 (2005).

[3] Engler, A.J., Sen, S., Sweeney, H.L. & Discher, D.E. Cell 126, 677-689 (2006).

[4] Guo, W.H., Frey, M.T., Burnham, N.A. & Wang, Y.-l. Biophysical Journal 90, 2213-2220 (2006).

[5] Gardel, M.L., Schneider, I.C., Aratyn-Schaus, Y. & Waterman, C.M. Annual Review of Cell and Developmental Biology 26, 315-333 (2010).

C.12 Reaction-diffusion models of bacterial *min* protein dynamics

Zdeněk Petrášek, Petra Schwille

Max Planck Institute of Biochemistry, Martinsried, Germany

The *min* proteins in rod-like *E. coli* bacteria interact with the inner membrane surface, where they organize into a dynamic pattern oscillating between the two cell poles. In an experimental model system consisting of a flat membrane with effectively infinite supply of proteins and energy source, the proteins assemble into travelling waves [1]. These experiments showed that only two proteins, the energy source and a lipid membrane of given composition are sufficent to produce the complex dynamic behaviour.
We explore models of the *min* system with interactions described by chemical kinetics allowing at most bi-molecular reactions, and a limited number of species. Our interest is to find out which minimal sets of interactions between the proteins and the membrane, compatible with the experimental biochemical evidence, can cause the observed spatial patterns and dynamics. We discuss which reaction schemes lead to unstable homogeneous steady states and limit cycles, and how diffusion affects the stability of these limit cycles. One of the goals is to determine the dependencies of the observable pattern parameters (period, wavelength) on the molecular interaction parameters (reaction rate constants, diffusion coefficients), and identify those that allow discrimination between alternative models by a quantitative comparison with experiments.

[1] M. Loose, E. Fischer-Friedrich, J. Ries, K. Kruse, and P. Schwille. Spatial regulators for bacterial cell division self-organize into surface waves in vitro. Science, 320(5877):789{792, 2008.

C.13 Tension-oriented cell divisions in zebrafish epiboly

Pedro Campinho[1], Martin Behrndt[1], Jonas Ranft[2,3], Thomas Risler[2], Nicolas Minc[4] and Carl-Philipp Heisenberg[1]

[1]*Institute of Science and Technology Austria, Klosterneuburg, Austria*

[2]*Laboratoire Physicochimie Curie, Institut Curie, Paris, France*

[3]*Laboratoire de Physique Statistique ENS, Paris, France*

[4]*Institut Jacques Monod, Paris, France*

Epithelial spreading is a common and fundamental aspect of various developmental and disease-related processes such as epithelial closure and wound healing. A key challenge for epithelial tissues undergoing spreading is to increase their surface area without disrupting epithelial integrity. Combining experimental and theoretical approaches, we show that orienting cell divisions by tension constitutes an efficient mechanism by which the enveloping cell layer (EVL) releases anisotropic tension while undergoing spreading during zebrafish epiboly. In the absence of tension-oriented cell divisions and in the presence of increased tissue tension, EVL cells undergo ectopic fusions, suggesting that the reduction of tension anisotropy by oriented cell divisions is required to maintain tissue integrity. Cell-division orientation by tension appears therefore as a key mechanism for promoting tissue spreading during EVL epiboly.

[1] P. Campinho, M. Behrndt, J. Ranft, T. Risler, N. Minc, and C.-P. Heisenberg, Nat. Cell Biol. 15 (12), 1405 (2013).

C.14 A simple active gel *in vivo*: the cytokinetic ring

Daniel Riveline

Laboratory of Cell Physics, ISIS/IGBMC, Strasbourg, France

Cell division is a fundamental event for single cells and multicellular organisms with implications in embryo development. At the onset of cytokinesis, a ring of actin filaments and myosin motors forms and constricts, and this leads to the separation of cells. Although molecular players are known, it is not yet understood how collective interactions of thousands of filaments and motors cause the ring closure. This mesoscopic question is our main focus with this unique system inside a cell: the cytokinetic ring is a complete active gel which can be entirely captured both experimentally and theoretically. We have designed an integratedapproach: we do experiments to probe the molecular interactions and dynamics of the ring in microfabricated chambers [1, 2, 3] and we develop physical models to which we compare our results. We apply this approach to cytokinetic rings in mammalian cell and in fission yeast. Both rings reveal new patterns and dynamics.

[1] Riveline, D. and Buguin, A. Devices and methods for observing the cell division. WO/2010/092116. Patent (2010).

[2] Riveline, D. and Wollrab, V. Devices and methods for observing eukaryotic cells without cell wall. WO/2013/135809. Patent (2013).

[3] Riveline, D. Methods for observing cells with cell wall or invertebrate embryos with oblong eggshell. WO/2013/144302. Patent (2013).

C.15 The mystery of circling microtubules: tubulin lattice switches under force

Falko Ziebert[1], Hervé Mohrbach[2], Igor M. Kulic[3]

[1]*Institute of Physics, Albert-Ludwig-University of Freiburg, Freiburg, Germany*

[2]*Groupe BioPhysStat, Université de Lorraine, Metz, France*

[3]*Institut Charles Sadron UPR22-CNRS, Strasbourg, France*

Eucaryotic cells do much rely on the mechanical properties of microtubules (MTs). Despite significant efforts, understanding the MTs unusual inner mechanics remained so far elusive. A persistent, to date unresolved mystery is the formation of gliding MT arcs and rings in kinesin-driven gliding assays, a phenomenon that seemingly contradicts any simple (e.g. worm-like chain) model of their mechanics. To elucidate these metastable curved states, we develop a novel model of the inner workings of the MT's lattice. It is based on experimental evidence for a distinct conformational switch of the tubulin dimer. The MT lattice itself can hence coexist in a number of discrete polymorphic states, which can be triggered by external mechanical loads. This lattice switch of the MTs in vitro, that remains concealed in the absence of forces but powerfully displays its existence in their presence, is expected to have important consequences in vivo.

[1] F. Ziebert, H. Mohrbach, I. M. Kulic, submitted (2014).

Poster Abstracts

P.1 ssDNA hybridisation in competition

Björn Ackermann

Biological Experimental Physics, Saarland University, Saarbrücken, Germany

We study the hybridization of many ssDNA oligonucleotides to surface bound probes. Our goal is to determine the largest possible number of different strands at a given length that hybridize in thermal equilibrium. We identify three different groups of sequences that are not useful due to unwanted conformations or tertiary structures. The first group consists of completely or partially self-complementary sequences. They will bind to each other in bulk, and they are less likely to bind to the surface bound probes. They even may even form beacons. The second group consists of sequences with runs of the same base, e.g. more than six times 'G' in one run GGGGGGG. We have shown that sequences with a length of 17 base pairs up to five mismatches interact. In future work we will study the case of interacting and competing, or non-competing sequences in more detail.

P.2 Inhibition of SOCE by ROS is mediated by direct oxidation and altered kinetics of Orai1 protein diffusion

Dalia Alansary[1], Martin Peglow[2], Heiko Rieger[2], Ivan Bogeski[1] & Barbara A. Niemeyer[1]

[1]*Department of Biophysics, Saarland University, Homburg, Germany*

[2]*Department of Theoretical Physics, Saarland University, Saarbrücken, Germany*

Store-operated Ca^{2+} entry (SOCE) is essential for T cell activation, differentiation and proliferation and is mediated by Orai ion channels in complex with activating endoplasmic reticulum (ER) membrane-resident STIM proteins. SOCE and I_{CRAC} mediated by Orai1, but not the homologue Orai3, are inhibited by reactive oxygen species. Upon antigen mediated T cell receptor stimulation, differentiation of naïve T cells into effector cells is accompanied by increased level of ROS within inflamed tissues and alteration of expression levels of Orai homologues in effector T cells in favor of the redox insensitive Orai3. Here we apply patch-clamp and microscopy approaches to further investigate the molecular mechanism of ROS dependent inhibition of I_{CRAC} mediated by Orai1 and STIM1 [1]. TIRF analysis shows that ROS inhibit the clustering rate of Orai1 WT but not of a mutant lacking all susceptible cysteine residues (TM). In addition, ROS are able to slow down WT but not TM diffusion as indicated by the longer time constants (τ) observed in FRAP measurements of WT compared to the TM. An Orai1 mutant C195D that mimics addition of charges at position 195 shows significantly reduced I_{CRAC} simulating wild type (WT) ROS treated channels, confirming that ROS inhibition is likely mediated by alteration of the charge at position 195. Furthermore, we are applying our mathematical tool to delineate the effects of altered STIM1 or Orai1 diffusion parameters from stoichiometrical changes [2]. Current investigations concern the dependence of charge and diffusion to understand how oxidation of Orai1 by ROS leads to slowed down diffusion, inefficient clustering and smaller currents.

[1] Bogeski I, Kummerow C, Al-Ansary D, Schwarz EC, Koehler R, Kozai D, Takahashi N, Peinelt C, Griesemer D, Bozem M, Mori Y, Hoth M, Niemeyer BA (2010) Differential redox regulation of ORAI ion channels: a mechanism to tune cellular calcium signaling. *Science Signaling*, 3, ra24.

[2] Kilch T, Alansary D, Peglow M, Dörr K, Rychkov G, Rieger H, Peinelt C, Niemeyer BA. (2013) Mutations of the Ca2+-sensing stromal interaction molecule STIM1 regulate Ca2+ influx by altered oligomerization of STIM1 and by destabilization of the Ca2+ channel Orai1. *J Biol Chem*. 18, 1653-64.

P.3 Intercellular communication between endothelial cells *via* tunneling nanotubes

Ksenia Astanina[1], Marcus Koch[2], Christian Jüngst[3], Andreas Zumbusch[3], Alexandra K. Kiemer[1]

[1]*Pharmaceutical Biology, Saarland University, Saarbrücken, Germany,* [2]*Leibniz Institute for New Materials, Saarbrücken, Germany,* [3]*Department of Chemistry, Konstanz University, Konstanz, Germany.*

Intercellular communication is one of the fundamental processes in the development and functioning of multicellular organisms. Known mechanisms of cell-to-cell interaction include exocytosis or direct transfer of small cytoplasmic components *via* gap junctions. Recently, an essentially new type of intercellular communication, based on thin membrane channels between mammalian cells, has been reported. These structures, called intercellular or tunneling nanotubes (TNTs) permit direct exchange of various components or signals (ions, proteins, organelles) between non-adjacent cells at distances over 100 µm. The functional role of this type of intercellular communication between endothelial cells has as yet not been investigated. Therefore, aim of this study was to characterize endothelial TNT morphology, cytoskeletal organization, and cargo.

Our studies show the presence of tunneling nanotubes in microvascular endothelial cells (HMEC-1). The TNTs were studied with live cell imaging, environmental scanning electron microscopy (ESEM), and Coherent Anti-stokes Raman scattering

Spectroscopy (CARS). Tunneling nanotubes showed distinct persistence: the TNTs could connect cells over long distances (up to 150 µm) for several hours. Even during mitosis dividing cells were connected to other cells by long TNTs. Plasma membrane staining revealed that the whole nanotube always belongs to one "host" cell. We observed at least three types of TNTs: either actin- or tubulin-containing TNTs or nanotubes with both actin filaments and microtubules. We have also studied the effects of stabilization/destabilization of cytoskeletal filaments on TNT length. Several cellular organelles were present in TNTs, for example lysosomes and mitochondria. Moreover, we could identify lipid droplets as a novel type of cargo in TNTs. Under angiogenic conditions (VEGF treatment) the number of lipid droplets increased significantly. Arachidonic acid application resulted not only in the increased number of lipid droplets, but also in TNT formation.
Taken together, our findings represent the first characterization of TNTs connecting HMEC-1 cells and report lipid droplets as a new type of TNT cargo.

P.4 Physics of membrane-cortex adhesion and bleb nucleation

Ricard Alert and Jaume Casademunt

Departament d'Estructura i Constituents de la Matèria, Universitat de Barcelona, Barcelona, Spain

We present a model for membrane-cortex adhesion. The model describes the dynamics of membrane undulations by coupling the nonlinear force-dependent adhesion kinetics to membrane elasticity and hydrodynamics. The spectrum and amplitude of membrane undulations under adhesion-induced correlations are analytically computed. These predictions could allow the fraction of connected membrane-cortex linker molecules to be experimentally determined from measurements of undulation spectra of cell membranes. Our model is also employed to study the nucleation of blebs. We find that the regime of membrane undulations responsible for spontaneous bleb

formation is controlled by adhesion rather than elastic properties of the membrane. This allows the cell to trigger or inhibit bleb formation via control of the activity of cortical myosin, with implications for directed bleb-based motility. The kinetics of bleb nucleation is studied via stochastic simulations of the continuum model with a consistent nucleation criterion.

P.5 Boundary integral formulation for Stokes flow: application to the flow of red blood cells

Othmane Aouane[1,2,3], Marine Thiébaud[2], Chaouqi Misbah[2], and Christian Wagner[1]

[1]Experimental Physics, Saarland University, Saarbrücken, Germany

[2]LIPHY, Université Grenoble Alpes, Grenoble, France

[3]LMPHE, URAC 12, Faculté des Sciences, Université Mohammed V- Agdal, Rabat, Morocco

We investigate numerically the flow of deformable objects such as vesicles and red blood cells (rbcs) in micro-channels. We focus on understanding the phenomena behind the formation of small train of cells (called clusters) that occurs in the microcirculation. We consider the rbc in 2D as a closed deformable and non-permeable membrane encapsulating an inner fluid and suspended in an outer fluid. The reduced area of the cell is taken to be that of a human red cell. The membrane total force is composed from a bending force, a tension force to fulfill the area conservation constraint, and a cell-cell interaction force to reproduce the depletion forces due to the effect of the plasma macromolecules. The cells are placed in a confined geometry (two parallel walls) and subjected to a Poiseuille flow. The boundary conditions could be resumed into: i) no slip condition, ii) stress balance, and iii) membrane inextensibility. The inner and outer fluids obey to Stokes equations. This equations are solved using the boundary integral formulation with the use of two walls green functions [1]. We observe that two kind of clusters exist namely: i) hydrodynamical clusters [2], and ii) polymer induced clusters [3]. We notice that the

hydrodynamical clusters are observed only when the confinement is too high or too low. For middle confinement, the hydrodynamical clusters do not exist. The polymer induced clusters are more robust and remain stable independently from the confinement.

[1] M. Thiébaud and C. Misbah, Phys. Rev. E 88, 062707 (2013).

[2] G. Tomaiuolo, L. Lanotte, G. Ghigliotti, C. Misbah, and S. Guido, Physics of Fluids (1994-present) 24, 051903 (2012).

[3] M. Brust, O. Aouane, M. Thiébaud, D. Flormann, C. Verdier, L. Kaestner, M. Laschke, H. Selmi, A. Beny- oussef, T. Podgorski, G. Coupier, C. Misbah, and C. Wagner, Scientific reports 4 (2014).

P.6 Membrane processes in biomineralization

Vera Bandmann, Marie-Louise Lemloh, Diana Peckys, Niels de Jonge and Ingrid Weiss

INM-Leibniz-Institute for New Materials, Biomineralization, Saarbrücken, Germany

A challenging goal in the understanding of controlled biomineralization is the uncovering of molecular mechanisms that lead to the self-organization of biological interfaces. Of special interest are certain intracellular processes for guiding the extracellular organization of biomaterials.

A myosin chitin synthase is one of the key enzymes regulating the organization of bio-minerals in mollusc. The pH dependent assembly of the extracellular domains of this transmembrane protein suggests that molecular cross-talk between the exterior and the interior of cells is a prerequisite for genetically controlled shell formation [1]. Strong evidence for the importance of intracellular transport of biomaterials was provided in the study of unicellular *Coleps hirtus* [2]. In these organisms, inorganic precursors for biomaterials are packed in vesicles, and are presumably transported to the mineralization site. Although various insights were obtained experimentally by electron microscopy of thin sections, important questions continue to remain puzzling. To understand how the organic and inorganic components of biominerals are stabilized, how the transport is controlled and where,

the crucial steps are located within the cell it would be desirable to employ methods capable of directly observing and locating these processes.
In this context, we discuss two experimental methods from different fields of research. High-resolution capacitance measurements allow the detection of the fusion and fission of single vesicles in real time [3]. Furthermore, nanoscale information of whole cells in liquid environment can be visualized using scanning transmission electron microscopy (STEM) [4]. Both methods provide a very high temporal and spatial resolution and experiments can be carried out close to *in vivo* conditions of the living cell. By combining both methods we aim to correlate intracellular trafficking, membrane dynamics and the extracellular organization of biological materials into hierarchical structures as formed by organisms in many different ways.

[1] Weiss, I. M., Lüke, F., Eichner, N., Guth, C., & Clausen-Schaumann, H. (2013). On the function of chitin synthase extracellular domains in biomineralization. Journal of Structural Biology, 183(2), 216–25.

[2] Lemloh, M.-L., Marin, F., Herbst, F., Plasseraud, L., Schweikert, M., Baier, J., … Brümmer, F. (2013). Genesis of amorphous calcium carbonate containing alveolar plates in the ciliate Coleps hirtus (Ciliophora, Prostomatea). Journal of Structural Biology, 181(2), 155–61.

[3] Bandmann, V., Kreft, M., & Homann, U. (2011). Modes of exocytotic and endocytotic events in tobacco BY-2 protoplasts. Molecular Plant, 4(2), 241–251.

[4] Peckys, D. B., Baudoin, J.-P., Eder, M., Werner, U., & de Jonge, N. (2013). Epidermal growth factor receptor subunit locations determined in hydrated cells with environmental scanning electron microscopy. Sci. Rep., 3, 2626.

P.7 Intragenic epigenetic modifications modulate exon usage in human across developmental stages

Ahmad Barghash, Siba Shanak, Volkhard Helms

Center for Bioinformatics, Saarland University, Saarbrücken, Germany

In this project we investigate the influence of epigenetic modifications on the mechanism of alternative splicing of messenger RNA and its

relationship to human development. Differential exon usage has been reported to affect several genes in mammalian genomes. Different splice variants can be linked to several regulatory domains of the same protein that can carry out several functions across different states. Recent studies investigated the significance of differential exon usage across several primate species in some adult tissues. They concluded that the effect is more profound across species than on the intra-species level. Other studies have attempted to recover the relationship between alternative splicing and epigenetic modifications. However, no study has addressed the issue how differential usage of exons is related to organismal development or to differential epigenetic modifications on the exon level. In this project, we study how differential exon usage is related to epigenetic modifications on the exon level, namely DNA methylation and several histone marks across different stages of human development. Furthermore, we checked for possible concurrent changes at the level of whole gene expression and the methylation state and histone marks. Associations are interpreted in terms of the enrichment of gene ontology terms for highly correlated genes.

[1] Reyes A, Anders S, Weatheritt RJ, Gibson, TJ, Steinmetz LM, Huber W (2013) Drift and conservation of differential exon usage across tissues in primate species. Proc Natl Acad Sci U S A. 110: 15377-15382.

[2] Zhou HL, Luo G, Jo Ann Wise JA, and Lou H (2013) Regulation of alternative splicing by local histone modifications: potential roles for RNA-guided mechanisms. Nucl. Acids Res. 42: 701-713.

P.8 The Xenopus spindle as an active liquid crystal

Baumgart J., Brugués J., Erlenkämper C., Joanny J.-F., Jülicher F.

Max Planck Institute for the Physics of Complex Systems, Dresden, Germany

In cells the spindle apparatus reliably segregates chromosomes. The main component are hundreds of thousands dynamic microtubules, which are well studied. Despite this detailed knowledge an understanding of how their collective properties give rise to a spindle with a defined size and shape is still lacking. To investigate how the

self-organization of the microtubules relates to a proper shaped spindle we use active liquid crystal theory. This approach is motivated by the rapid turnover of microtubules, their steric interactions, and the motor proteins acting between pairs of microtubules. Such a description allows us to analyze spindle mechanics by nematic field equations in an active fluid. Here we study a description including the orientation field, active stesses due to motors, and a source term to account for microtubule nucleation. We ignore convective terms and coupling of the flow field on the orientation field. In two dimensions we solve the set of linear differential equations by means of the finite element method.

P.9 KDEL receptor dynamics, internalization and retrograde transport in mammalian and yeast cells

Björn Becker, Domenik Rammo and Manfred J. Schmitt

Molecular & Cell Biology, Department of Biosciences, Saarland University, Saarbrücken, Germany

A/B toxins such as cholera toxin and the yeast K28 toxin possess an H/KDEL amino acid motif at the C-terminus of their cell binding B-subunit which is recognized and bound by KDEL receptors (KDELRs) of their target cells. The main function of KDELRs is to retrieve resident ER proteins from the late secretory pathway back to the ER. Until now, it was believed that the initial toxin interaction with KDELRs occurs within the Golgi, i.e. after receptor-mediated endocytosis and endosomal trafficking. However, we recently demonstrated that the yeast KDEL receptor (Erd2p) is not only present in the ER and Golgi but also in the plasma membrane (PM). We demonstrate that PM localized yeast KDELR is capable to bind Kar2p/BiP from outside the cell, leading to its endocytosis and retrograde transport to the ER where it restores cell growth of a *kar2*ts mutant. In addition to its essential physiological role in the uptake of secreted H/KDEL-bearing proteins from the PM, we show that Erd2p is crucial for binding and endocytosis of the viral

A/B toxin K28 in yeast. To understand the mechanistic principles of spatio-temporal dynamics, compartmentalization and clustering of KDELRs in yeast and higher eukaryotic cells, we are using fluorescently labelled variants of both, KDELRS and model cargo (K28, ricin).
By using confocal laser scanning microscopy, live cell imaging, colocalization studies and *in vivo* toxicity assays, we are able to visualize toxin internalization as well as receptor dynamics in mammalian cells. Thereby, time-dependent toxin/receptor clustering at the plasma membrane can only be observed with cargo containing a C-terminal ER retention motif. We are currently working on a mathematical model that will help us to describe the process of toxin/receptor cluster formation at the cell surface. Moreover, we perform live cell imaging of fluorescent KDEL receptor variants in order to dissect and quantify compartmental distribution and dynamics of KDELRs in living cells.

P.10 TRPC channels mediate Ca^{2+} influx and migration of astrocytes

Belkacemi, Thabet; Philipp, Stefan E.; Weißgerber, Petra; Flockerzi, Veit; Beck, Andreas

Institut für Experimentelle und Klinische Pharmakologie und Toxikologie, Saarland University, Homburg, Germany

After an acute brain injury a severe astrogliosis, i.e. increased proliferation and migration of astrocytes, occurs in the injured brain region. Proliferation and migration are triggered by cytosolic Ca^{2+} signals, due to intracellular Ca^{2+} release and/or Ca^{2+} influx. Canonical transient receptor potential channels (TRPCs) and TRPA1 channels are non-selective Ca^{2+} permeable ion channels, and suggested to be expressed in astrocytes. They might be involved in the glial Ca^{2+} signaling and thereby in migration and proliferation.
Using a fluorescence-labeled antibody against the astrocyte-specific surface marker glutamate aspartate transporter (GLAST), we sorted mouse cortical astrocytes, derived from 1-3 days old pubs and cultured for two weeks, by fluorescence-activated cell sorting (FACS). From 5

independent cell preparations RNA was prepared to perform RT-PCR for TRPC transcripts. Consistently, TRPC1-, TRPC2-, TRPC3- and TRPC4 cDNA fragments were amplified. Next we used various agonists to induce Ca^{2+} entry. Calcium imaging experiments, using the Ca^{2+}-sensitive fluorescent dye Fura-2, revealed OAG-mediated Ca^{2+} entry and intracellular Ca^{2+} oscillations in astrocytes isolated from wild-type mice. OAG, an analogue of diacylglycerol (DAG), is known to activate TRPC2, TRPC3, TRPC6 and TRPC7. We used astrocytes from various TRPC-deficient mouse strains to delineate the TRPC channel responsible for OAG-induced Ca^{2+} signaling. In addition we performed in vitro migration assays of wild-type and TRPC-deficient astrocytes, and studied glial scar formation after inducing cortical stab lesions. The results presented show that TRPC-dependent Ca^{2+} signaling is important in astrocyte migration, and probably in astrogliosis after severe brain injury.

P.11 Chemosensory responses of Grueneberg Ganglion Neurons depend on CNGA3

Katherin Bleymehl, Trese Leinders-Zufall, Frank Zufall

University of Saarland, School of Medicine, Department of Physiology, Homburg, Germany

Neurons of the Grueneberg Ganglion (GGNs) are located at the rostral tip of the nose and project to glomeruli in the caudal region of the olfactory bulb. They have been described to detect cold temperatures (Mamasuew et al. 2008 and Schmid et al. 2010) and some odor ligands (Brechbühl et al. 2008, Mamasuew et al. 2011), including alarm pheromones. Like all other mature olfactory neurons they express olfactory marker protein (OMP). In addition, various chemsensory receptors, like TAAR´s and V2R2 (Fleischer et al. 2006 and Fleischer et al. 2007), as well as elements of a cGMP (Fleischer et al 2009 and Liu et al 2009) second messenger pathway have been described in these neurons. Yet, the molecular mechanisms underlying the odor- induced responses are still far from clear.

To determine the dependence of chemosensory Grueneberg ganglion (GG) responses on the presence of cyclic nucleotide-gated ion channels CNGA3 and to the Guanylate cyclase type G (GCG) we recorded Ca^{2+} signals from GGNs in coronal GG tissue slices freshly prepared from male and female OMP-GFP$^{+/-}$, CNGA3$^{-/-}$ OMP-GFP$^{+/-}$ and GCG$^{-/-}$ OMP-GFP$^{+/-}$ mice. To enable us to monitor intracellular Ca^{2+} changes in GGN somata, we loaded the slices with the Ca^{2+} indicator Fura-2/AM. The GGN identification occurs due to their expression of GFP. This permitted us to perform simultaneous Ca^{2+} measurement on identified GFP-labelled GGNs using Fura-2. GGNs produced robust and reproducible increases in Ca^{2+} when stimulated with 2,3 – dimethylpyrazine (2,3-DMP), 2 – propylthiethane (2-PT), 2,4,5 trimethylthiazoline (TMT) and other chemoligands. The responsiveness of GGNs to the various ligands was then investigated in mice deficient for CNGA3 or GCG. In fact in CNGA3 deficient mice the response frequency is reduced, suggesting the involvement of cGMP in the detection of these chemoligands. GCG seems not to be involved in the signal transduction pathway of TMT and the DMP-induced responses. However, 2-PT responses occur at a lower frequency similar to the measurements in CNGA3-deficient mice suggesting different signal transduction pathways in GGNs.

P.12 The role of ORAI channels and their interplay with NADPH oxidase 2 in bacterial peptide-induced innate immune responses

Stephanie Saul, Reinhard Kappl, David Conrad, Bastian Pasieka, Carsten Kummerow, Barbara Niemeyer, Ivan Bogeski

Department of Biophysics, School of Medicine, Homburg, Germany

One of the major events during phagocytosis is recognition of bacterial peptides by Formyl peptide receptors (FPRs); an event which is followed by Ca^{2+} mobilization and production of reactive oxygen species (ROS) via the NADPH-oxidase (NOX) enzymes. We have

previously shown that oxidation reduces store operated Ca^{2+} entry (SOCE) by inhibiting ORAI1 and 2 but not ORAI3 channels and identified upregulation of ORAI3 expression as an adaptive mechanism against oxidative stress. However, the roles of ORAI channels and their activators STIM1 and STIM2 and the molecular mechanisms by which they might regulate NOX activity, and vice versa, are still not fully understood. Here, we show that besides ORAI1 and STIM1, primary human monocytes express relatively high amounts of ORAI3 and STIM2. The ORAI1/ORAI3 expression ratio correlates with the redox sensitivity of SOCE as well as with the viability of the human monocytes under oxidative stress. Furthermore, by patch-clamp recordings of ORAI1-ORAI3 concatamers we show that a single subunit of ORAI3 renders the ORAI multimers redox insensitive. Moreover, electron paramagnetic resonance (EPR) and fluorescent ROS measurements indicate that primary human monocytes generate superoxide ($O2^{\bullet-}$) upon activation of SOCE either by FPR activation by bacterial peptides or by passive store depletion. This SOCE-induced $O2^{\bullet-}$ production was significantly reduced by the NOX inhibitor DPI, under Ca^{2+}-free conditions and upon silencing of ORAI1, STIM1 or STIM2, suggesting a major role for SOCE in the regulation of NOX2 activity. To test the role of the redox-insensitive ORAI3 in the increased redox resistance of human monocytes, we silenced ORAI3 expression using small interfering RNA. Measurements of SOCE and ROS production showed increased oxidation-induced inhibition of NOX2 activity in ORAI3 silenced monocytes. In summary, we demonstrate existence of a regulatory feedback loop between ORAI channels and NOX2 enzymes which might be involved in prevention of phagocyte-induced host tissue damage.

P.13 Formation of long lived local protein kinase C clusters after short Calcium puffs

Mike Bonny[1], Xin Hui[2], Peter Lipp[2] and Karsten Kruse[1]

[1]*Theoretical Physics, Saarland University, Saarbrücken, Germany*

[2]*Institute for Molecular Cell Biology, Saarland University, Homburg, Germany*

Conventional protein kinases C (cPKCs) play an important role in signal transduction and in gene regulation. PKCα, a member of the cPKC-family, translocates to the plasma membrane after activation via cytosolic Ca^{2+} ions. In particular, there exist local translocation events, when PKCα forms clus- ters on the membrane with limited spatial spreads ($< 4\mu m$). The lifetime of brief events is 400-1500ms, while long lasting events have a lifetime larger than 5s, which markedly exceeds the duration of a Ca^{2+} puff [Reither et al., 2007]. We show theoretically that allosteric effects together with interactions between membrane-bound PKCα molecules can lead to the observed behaviour.

P.14 Clustering of red blood cells in microcapillaries under flow conditions

Viviana Clavería[1,2], Manouk Abkarian[1] and Christian Wagner[2]

[1]*Laboratoire Charles Coulomb, Univ. Montpellier 2, Montpellier, France*

[2]*Experimental Physics, Saarland University, Saarbrücken, Germany*

Red blood cells (RBCs) can interact at rest and form aggregates called rouleaux due to the presence of macromolecules in the surrounding media. Under macroscopic flow, the rouleaux are broken up again and hence one might speculate that they not play a role in the physiological blood flow [1]. Nevertheless, it has been reported that the presence of fibrinogen or dextran leads to an enhanced formation of robust clusters

of red blood cells under flow conditions in microcapillaries [2]. It has been also report that consecutive RBCs in microcapillaries tend to associate into clusters attributing the effect to hydrodynamics interactions between the cells [3]. Despite the observations in vivo [2], in vitro and numerical simulations [4], the physics behind this mechanism is still unclear.
We will present systematic experimental research on cluster formation of RBCs on microcapillaries under flow conditions. In order to mimic the flow through the microvasculature, a microfluidic device with narrow channels with diameters in the order of a RBC diameter at rest was designed. The experimental methods are mostly flow visualization and image treatment including particle tracking in order to characterize the number of clusters, the size of the cluster in terms of number of cells and spatial size and the form of the red blood cells in the flow. Our controlling variables are the hematocrit level inside the microchannels, the pressure drop of the imposing flow and dextran concentration in the suspending media.

[1] Popel, Aleksander S., and Paul C. Johnson. "Microcirculation and hemorheology." Annual review of fluid mechanics 37 (2005): 43.

[2] Brust, Matthias, et al. "The plasma protein fibrinogen stabilizes clusters of red blood cells in microcapillary flows." Scientific reports 4 (2014).

[3] Tomaiuolo, Giovanna, et al. "Red blood cell clustering in Poiseuille microcapillary flow." Physics of Fluids 24.5 (2012): 051903.

[4] Ghigliotti, Giovanni, et al. "Why and how does collective red blood cells motion occur in the blood microcirculation?." Physics of Fluids (1994-present) 24.10 (2012): 101901.

P.15 Managing oral biofilm formation with plant extracts

Sarah Colanesi, Xiaoting Jin, Natalia Umanskaya and Matthias Hannig

Clinic of Operative Dentistry, Periodontology and Preventive Dentistry,Saarland University, Homburg, Germany

Conventional management of oral biofilm formation is based on daily oral hygiene (tooth brushing and flossing) as well as use of mouth rinsing solutions (e.g. chlorhexidine). Here, we present a novel

approach to control bacterial adherence and oral biofilm formation by application of mouth wash solutions containing compounds from plant extracts.
Enamel specimens were mounted to intraoral splints and carried by up to five volunteers over 24 h for in situ biofilm formation. Two times rinsing with experimental mouth wash solutions containing among other ingredients peppermint oil and rosemary oil took place after 3 min and 12 h of intraoral exposure. After removal from the splints, the specimens were analysed by fluorescence microscopy, scanning and transmission electron microscopy.
Microscopic analyses demonstrated that 24-h in situ biofilm formation was significantly reduced due to two times rinsing with the experimental solutions compared to control specimens without rinsing. Fluorescence microscopic evaluation additionally showed a lower viability of the adherent bacteria after rinsing with the plant extract containing solutions compared to controls.
Mouth rinsing solutions containing plant extracts reveal an interesting approach to influence the process of bacterial adherence and to control bacterial biofilm formation under oral conditions.

P.16 A mechanistic understanding towards cellular uptake

S. Dasgupta, T. Auth, G. Gompper

Institute of Complex Systems and Institute for Advanced Simulation, Forschungszentrum Jülich, Jülich, Germany

Budding of cell membranes initiates intracellular vesicle transport and has been studied for a variety of soft matter systems. Using a continuum model, we study wrapping of a single nano-particle as an interplay of the membrane deformation energy and the adhesion energy of the particle and membrane. With the help of numerical energy minimization using triangulated surfaces, we investigate the role of shape and size of the particle as well as of the membrane's elastic parameters on nano-particle wrapping.
For rod-like and disc-like particles, we find a higher binding affinity to the membrane compared with spherical particles. However, such particles have a lower uptake to cells, as confirmed by experiments. All

more complex particle shapes, such as a Hauser's cube and supereggs, have stable partially-wrapped states with shallow and high wrapping fractions for sufficiently high adhesion strengths. Partially-wrapped particles can be advantageous both from an application point of view as well as from a biological point of view. Particles can also preferentially bud tip first from the membrane similar to filamentous viruses like Ebola and Marburg.
Similarly, a mechanistic understanding of how the malarial merozoite invades the erythrocyte membrane tip-first is lacking. Assuming an asymmetric egg-like shape for the parasite, we have investigated the role of the different membrane properties on the invasion mechanism. We propose a concentration gradient of adhesive molecules to be responsible for the reorientatioin of the particle towards the tip before invasion and membrane spontaneous curvature from cytoskeletal re-modelling or secretion of unstructured membranes to assist motor forces for the parasite to invade a red blood cell.

[1] S. Dasgupta, T. Auth and G. Gompper, Wrapping of ellipsoidal nano-particles by fluid membranes, Soft Matter, 9, 5473-5482, 2013.

[2] S. Dasgupta, T. Auth and G. Gompper, Wrapping of ellipsoidal nano-particles by fluid membranes, Nano Letter, 14, 687–693, 2014.

[3] S. Dasgupta et al., Erythrocyte membrane-wrapping contributions to malaria parasite invasion of the human erythrocyte (Manuscript in submission).

P.17 Scanning transmission electron microscopy of whole eukaryotic cells in liquid

Niels de Jonge and Diana B. Peckys

INM, Leibniz Institute for New Materials, Innovative Electron Microscopy, Saarbrücken, Germany

Scanning transmission electron microscopy (STEM) of samples in liquid, so-called Liquid STEM [1, 2], opens up the possibility of direct mapping protein complexes in intact eukaryotic cells in their native aqueous environment with nanometer resolution. The principle is as follows. Live eukaryotic cells are directly grown on silicon microchips

with silicon nitride (SiN) membrane windows, and then incubated with protein labels consisting of gold nanoparticles or fluorescent quantum dots. The cells are then fixed, and imaged in hydrated state with STEM. A resolution of 4 nm was achieved on nanoparticle labels for a water thickness of 6 μm [1]. The high resolution through such thick liquid layers is explained on the basis of the atomic number contrast of dark field STEM. It is also possible to image native (unfixed) cells [3]. Alternatively, environmental scanning electron microscopy (ESEM) with STEM detection can be used for thin cellular regions [4]. Liquid STEM was used to determine the stoichiometry of membrane protein complexes directly on whole cells.

[1] N. de Jonge et al, Proc. Natl. Acad. Sci. 106 (2009), 2159-2164.

[2] N. de Jonge and F.M. Ross, Nat. Nanotechnol. 6 (2011), 695-704.

[3] D. B. Peckys et al, Biophys. J. 100 (2011), 2522-2529.

[4] D. B. Peckys et al, Sci. Reports 3 (2013) 2626-1-6.

P.18 Biogenesis of large dense core vesicles in the adrenal chromaffin cells of newborn mice

Ekta Dembla and Ute Becherer

Department of Physiology, Saarland University, Homburg, Germany

Chromaffin cells are neuroendocrine cells which secrete catecholamines and peptides and thus play an important role in stress induced responses. The hormones that they release in a Ca^{2+} dependent manner are stored in large dense core vesicles (LDCVs) with a diameter of about 120 nm. Although adrenal chromaffin cell have served as a model system to study fast Ca^{2+}-dependent exocytosis for many years, very little is known about the biogenesis of LDCVs or their recycling. It has been shown that LDCVs are generated at the level of the trans Golgi network (TGN) under the control of chromogranins. Much less is known about sorting of associated and integral membrane components to the membrane of LDCVs.

In order to follow the biogenesis of LDCVs in chromaffin cells, we transfected the chromaffin cells with NPYmCherry, fixed them with

increasing delay and immunolabelled with cis-Golgi and plasma membrane marker. We found that LDCVs were retained at Golgi for one hour before moving through the cytoplasm to reach the plasma membrane. We also studied the association of vSNARE proteins Synaptobrevin-2 and Cellubrevin with LDCVs and found that Synaptobrevin-2 and Cellubrevin gets associated to LDCVs at a late stage of LDCV biogenesis. This indicates that there might be the fusion of precursor vesicles containing v-SNARE proteins with the LDCVs after they leave Golgi. Further, we are studying the endocytic and recycling pathway of LDCV's membrane components to get more insight into their fate.

P.19 v-SNARE-based protein-lipid interactions in exocytosis

M. Dhara, A. Yarzagaray, Y. Schwarz, R. Mohrmann, D. Bruns

Department of Physiology, Saarland University, Homburg, Germany

Regulated exocytosis is defined as the Ca^{2+}-triggered fusion of vesicular structures with the plasma membrane, enabling the release of cargo molecules. The fusion process involves a number of energetically complex steps that require both, protein-protein as well as protein-lipid interactions. We intend to experimentally address the nature of the interplay between synaptobrevin II (sybII) and phospholipids that seems crucial for Ca^{2+}-triggered neurotransmitter release.
Using a combination of photolytic 'uncaging' of intracellular Ca^{2+} with membrane capacitance measurement in chromaffin cells, we demonstrate that reduced flexibility of the sybII transmembrane domain (TMD) severely impairs Ca^{2+} triggered exocytosis, whereas mutants which show enhanced or similar flexibility to sybII can rescue secretion like the wild type protein.
Analysis of single amperometric spikes in combination with simultaneous membrane capacitance measurements reveal that reduced flexibility of the sybII TMD slows the kinetics of neurotransmitter discharge from single vesicle and reduces the fusion pore jitter. In contrast, TMD mutants with higher flexibility speed up

the kinetics of single vesicle fusion and enhance the fusion pore dynamics.
Thus our results demonstrate that SNARE TMDs play an active role in the fusion process that goes beyond simple anchoring of the protein. Specifically, we show that flexibility of TMD determines the magnitude of Ca^{2+} triggered exocytosis and kinetics of cargo discharge from single vesicles.

P.20 Spiral actin-polymerization waves can generate amoeboidal cell crawling

A. Dreher[1], I.S. Aranson[2] and K. Kruse[1]

[1]*Theoretical Physics, Saarland University, Saarbrücken, Germany*

[2]*Materials Science Division, Argonne National Laboratory, Argonne, USA*

Amoeboidal cell crawling on solid substrates is characterized by protrusions that seemingly appear randomly along the cell periphery and drive the cell forward. For many cell types it is known that the protrusions result from polymerization of the actin cytoskeleton. However, little is known about how the formation of protrusions is triggered and whether the appearance of subsequent protrusions is coordinated. Recently, the spontaneous formation of actin polymerization waves was observed. These waves have been proposed to orchestrate the cytoskeletal dynamics during cell crawling. Here, we study the impact of cytoskeletal polymerization waves on cell migration using a phase-field approach. In addition to directionally moving cells, we find states reminiscent of amoeboidal cell crawling. In this framework, new protrusions are seen to emerge from a nucleation process generating spiral actin waves in the cell interior. Nucleation of new spirals does not require noise, but occurs in a state that is apparently displaying spatio-temporal chaos.

P.21 Spontaneous Ca^{2+} signals in the mouse inner ear during development

Tobias Eckrich, Kerstin Blum, Stefan Münkner, Jutta Engel

Department of Biophysics, Saarland University, Homburg, Germany

Mammalian Inner hair cells (IHCs) are the sensory cells that transduce mechanical stimuli (sound) into graded receptor potentials leading to neurotransmitter release. Together with supporting cells they form the organ of Corti. During development, both inner hair cells and supporting cells undergo intricate morphological and physiological changes. Before the onset of hearing, they are transiently generating spontaneous Ca^{2+} based activity, which is thought to drive various developmental programs. IHCs generate Ca^{2+} action potentials based on voltage gated Ca^{2+} channels, whereas supporting cells exhibit Ca^{2+} waves spreading across the sensory epithelium through a reaction diffusion mechanism.

Live Ca^{2+} imaging using Fluo-8 AM was performed to characterize Ca^{2+} waves and transients in acute preparations of the mouse organ of Corti. Spontaneous Ca^{2+} transients of supporting cells with rise times of several seconds initiated Ca2+ waves, which then spread across the supporting cell epithelium at 5-7 µm/s. For the first time, Ca^{2+} transients were observed within the IHC region as well. These were limited to single cells and showed 100fold faster rise times.

Currently we are defining the molecular players that generate or modulate the fast Ca^{2+} transients and their interplay with the supporting cell Ca^{2+} waves using pharmacological tools and mouse models.

P.22 Interaction of bilayers in microfluidics: shedding light on the fast formation of single hemifused state

Nabor Jose Vargas[1], Ralf Seemann[1,2] and Jean-Baptiste Fleury[1]

[1]*Experimental Physics, Saarland University, Saarbrücken, Germany*

[2]*Max Planck Institute for Dynamics and Self-Organization, Göttingen, Germany*

Supported planar lipid bilayers generated by the classical Montal-Mueller (MM) method have been used for decades as model system to study the functional properties of ion channels and pores. This classical method suffers from several drawbacks since the supported Montal-Mueller planar bilayers are delicate and metastable structures with a short life time in an artificial geometry. Recently, a new method was proposed by H. Bayley et al.[1] which allows producing very stable planar lipid bilayer by contacting two water droplets in a surrounding oil phase (droplet interface bilayer). This approach provides an improved platform to measure ionic transport properties of membrane proteins in a more realistic non-supported environment.
Based on the concept of droplet interface bilayer we present an original and quasi automated microfluidic scheme which is able to produce and to study the formation of a single hemifused state between two free standing lipid bilayers. In a first step, two lipid bilayers are prepared at desired location from lipid decorated oil/water interfaces. In a second step, the two model membranes are brought in contact and form a single hemifused state within milliseconds, i.e. several orders of magnitude faster than reported in literature. The formation dynamics of the hemifused diaphragm is studied by simultaneous optical and electrophysiological measurements for four different lipids molecules. For all tested lipids, we could demonstrate that the formation of the hemifused state is a two-step process, where the second state is a dewetting process with no-slip boundary conditions. The resulting hemifused state is long living and we show the possibility to trigger a single fusion event by electrofusion [2].

[1] H. Bayley, B. Cronin, A. Heron, M. A. Holden, W. L. Hwang, R. Syeda, J. Thompson and M. Wallace, Mol. BioSyst., 2008, 4, 1191–1208.

[2] N. J. Vargas, R. Seemann and J-B Fleury (Submitted, 2014).

P.23 How to Study a Single SNARE Mediated Membrane Fusion Event ?

Jose Nabor Vargas[1], Kevin Howan[2], Ralf Seemann[1,3], Jean-Baptiste Fleury[1] and Frédéric Pincet[2,4]

[1]*Experimental Physics, Saarland University, 66123 Saarbrücken, Germany*

[2]*Laboratoire de Physique Statistique, Ecole Normale Supérieure, 75005 Paris, France*

[3]*Max Planck Institute for Dynamics and Self-Organization, 37077 Göttingen, Germany*

[4] *Department of Cell Biology, School of Medicine, Yale University, CT 06520 New Haven, USA.*

We present an approach to explore the properties of a single SNARE mediated membrane fusion event in a microfluidic chip. In a first step, a single free standing lipid membrane is generated at a defined position with the Droplet Interface Bilayer technique (DiB) [1]. In a second step, we inject a solution of divalent cations (Calcium, Ca2+) and small unilamellar vesicles functionalized with T-SNARE proteins (T-SUVs) around the planar membrane using a volume controlled flow. The presence of calcium mediates the direct fusion of the vesicles with the planar membrane, which is incorporating the proteins into the membrane. In a third step, we remove the calcium and the T-SUVs with a buffer solution. After this washing step, a solution of small unilamellar vesicles functionalized with V-SNARE proteins (V-SUVs) is injected around the planar membrane. And finally, we study single fusion event with good optical and electrical access.

[1] H. Bayley, B. Cronin, A. Heron, M. A. Holden, W. L. Hwang, R. Syeda, J. Thompson and M. Wallace, Mol. BioSyst., 2008, 4, 1191–1208.

P.24 Aggregation of red blood cells

Daniel Flormann and Christian Wagner

Dep. Experimental Physics, Saarland University, Saarbrücken, Germany

Plasma proteins such as fibrinogen or synthetic polymers such as dextran are well known as proteins or macromolecules with a strong influence on the red blood cell (RBC) aggregation. These aggregates, called rouleaux, are reversible and they are build and break up in the human body permanently. The presence of aggregated RBCs in a solution change the behavior of suspension significantly. The goal of our study to understand the physical mechanism of rouleaux formation. Macroscopic methods like sedimentation rates and rheometry show a similar behaviour to microscopic measurements (for example adhesion measurements with AFM). Still, there exist two opposing theories to describe the aggregation process (depletion or bridging). Our investigations suggest, that the depletion theory is more likely. Finally we clarify a controversial debate on the effect of the c-reactive (CRP) protein on the erythrocyte sedimentation rate (ESR). Several medical publications correlate the CPR concentrations in the human plasma with the ESR, which increase with increasing aggregation of RBCs. However, we could unambiguously show, that there is no relation between the CRP and the ESR without using any other plasma proteins.

P.25 The flagellar counterbend phenomenon

Hermes Gadelha[1], Eamonn. A. Gaffney[1] and Alain Goriely[2]

[1]Wolfson Centre for Mathematical Biology, and [2]Oxford Centre for Collaborative Applied Mathematics, Mathematical Institute, University of Oxford, Oxford, United Kingdom

Recent observations of flagellar counterbend in sperm show that the mechanical induction of curvature in one part of a passive flagellum induces a compensatory counter-curvature elsewhere. This apparent paradoxical effect cannot be explained using the standard elastic rod

theory of Euler and Bernoulli, or even the more general Cosserat theory of rods. Here, we develop a mechanical model capable of predicting the curvature reversal events observed in eukaryotic flagella. This is achieved by allowing the interaction of deformations in different material directions, by not only accounting for structural bending, but also the elastic forces originating from the cross-linking mechanics. Large amplitude configurations can be described analytically and an excellent match between the model and the observed counterbend deformation was found. This allowed a simultaneous estimation of multiple sperm flagellum material parameters, namely the cross-linking sliding resistance, the bending stiffness and the sperm head junction compliance ratio. Our analysis demonstrates that the counterbend emerges as a fundamental property of sliding resistance, which also suggests that cross-linking proteins may contribute to the regulation of the flagellar waveform in swimming sperm via counterbend mechanics. Finally, we investigate how the counterbend-type dynamics in sperm flagella is affected by viscous dissipation.

[1] Pelle DW, Brokaw CJ, Lesich KA & Lindemann CB, *Cytoskeleton* **66** 721735 (2009).

[2] Gadelha H, Gaffney EA & Goriely A, PNAS **110** (30), 12180 (2013).

P.26 Mechanism of Ca^{2+} leak from endoplasmic reticulum

Gamayun, I.[1], Schorr, S.[2], Dudek, J.[2], Zimmermann, R.[2], Cavalié, A.[1]

1Department of Experimental and Clinical Pharmacology &Toxicology

[2]Department of Medical Biochemistry & Molecular Biology, Saarland University, Homburg, Germany

The Ca^{2+} gradient between the endoplasmic reticulum (ER) lumen and the cytosol is maintained by the activity of SERCA pumps, while the numerous amounts of channels on the ER membrane allow Ca^{2+} leak to the cytosol. Sec61 complex is an important component of the translocon that mediates the transport of nascent proteins into the ER.

Growing evidence suggests that Sec61 additionally can also form a non-selective ionic channel that supports Ca^{2+} leak from the ER. We address the question under which condition Sec61 channels support Ca^{2+} leak. To this purpose, we have created a cell line of HEK293 cells stably expressing Ca^{2+} sensitive chameleon D1ER to provide FRET-based measurements of the Ca^{2+}-concentration in the ER. In our study we are using protein synthesis inhibitors (*Puromycin, Pactamycin, Cyclohexamide, Eeyarestatin1*) to show an impact on the Ca^{2+} leak induced by the application of irreversible blocker of SERCA pumps *Thapsigargin*. With simultaneous measurements of cytosolic Ca^{2+} (Fura 2) and lumenal Ca^{2+} (FRET), we have revealed the dose-dependent effect on the Ca^{2+} leak of these protein synthesis inhibitors on the *Thapsigargin*-induced depletion of the ER Ca^{2+}. Furthermore, both application of *Pactamycin* and *Eeyarestanin1* led by itself to the depletion of the ER. These experiments might provide insights on the mechanisms of Ca^{2+} leak associated to the protein synthesis.

P.27 Regulation of the cation channel protein Yvc1 by SUMOylation

J. Gartmann

Inst. f. Medical Biochemistry, Saarland University, Homburg, Germany

The yeast vacuolar conductance protein -Yvc1- belongs to the large family of evolutionary conserved *transient receptor potential* (TRP) cation channel proteins. It is located in the vacuolar membrane and is functionally involved in the homeostasis of ion concentration and osmoregulation in *Saccharomyces cerevisiae.* A single Yvc1 subunit contains six predicted transmembrane domains, two large cytoplasmic N- and C-terminal domains, and a linker region separating the fifth and sixth transmembrane domain. Four Yvc1 subunits are thought to constitute a tetrameric cation channel with their linker regions forming the ion conductance pathway. Since *Saccharomyces cerevisiae* is an ideal model organism for studying biological processes in eukaryotes, a closer investigation of Yvc1 is an easy approach to get more insights into the functions of the TRP channel protein family.

The small ubiquitin-like modifier (SUMO) is a posttranslational protein modification covalently attached to a lysine residue of its target protein. While in mammalian cells four different SUMO proteins regulate cellular processes, only one, Smt3, is necessary in *Saccharomyces cerevisiae.* Activation and conjugation of the SUMO protein as well as ligation to the target constitute the steps of a SUMO cycle. We found a link between Yvc1 and SUMO and investigate the regulation of Yvc1´s function by SUMOylation.
In vitro SUMOylation assays using recombinant proteins demonstrated that both Yvc1-N and Ycv1-C can be modified by SUMO. Analysis by mass spectrometry confirmed the interaction of Yvc1 and SUMO. In further studies, we aim to detect the specific SUMOylation sites of Yvc1 and investigate the effect of Yvc1 SUMOylation *in vivo*.

P.28 Decoding of processes influencing the (in)stable copying of DNA methylation patterns in mammalian cells

Pascal Giehr[1], Charalampos Kyriakopoulos[2], Jörn Walter[1], Verena Wolf[2]

[1]Institute of Epigenetics and [2]Computer Science Department, Saarland University, Saarbrücken, Germany

DNA methylation is an epigenetic modification associated with the regulation of gene function. Gene specific DNA-methylation patterns change over development and in specific cell types. DNA methyltransferases (Dnmts) in cooperation with other enzymes are responsible for the setting and maintenance of methyl groups at CpG-dinucleotides during cell division. The control of DNA methylation is highly dynamic involving regulated and random processes. The recent discovery of oxidative forms of DNA-methylation challenges some of the earlier proposed roles and concepts of DNA modifications. We combined a recently develop method, Oxidative Bisulfite Sequencing, with Hairpin Bisulfite Sequencing, allowing us to detect 5-methylcytosine as well as 5-hydroxymethyl cytosine of both DNA strands of one individual chromosome. By that we gain a closer look

into the dynamics of methylation and demethylation processes of dividing and non-dividing cells. By analyzing single copy genes and dispersed repetitive elements across the genome (LINE1, B1, IAP-LTR-retrotransposons, and major satellites) of embryonic stem cells cultivated on two different media we obtain a comprehensive picture of methylation dynamics at various regions in true genome of cells. Using this Data in a (hidden) Markov model we will be able to describe the evolution of methylation pattern within cells and decipher the role of the different enzymes of the methylation and demethylation machinery.

P.29 Intermittent motion of motor proteins in biological environments

Anne E. Hafner, M. Reza Shaebani, and Ludger Santen

Theoretical Physics, Saarland University, Saarbrücken, Germany

Motor proteins exhibit different types of anomalous diffusive behavior depending on whether or not they attach to cytoskeletal filaments. While they experience a subdiffusive dynamics within the cytoplasm due to a crowded environment, they usually move superdiffusively at short time scales due to the active transport on the filament network, followed by a normal diffusion at long times. Here we study an intermittent motion, composed of two different modes of motility, which mimic the properties of the biological environment. By means of extensive Monte Carlo simulations we investigate how the overall transport properties depend on the structural properties of the filament network, the stepping strategy in the crowded cytoplasm, and the motor processivity. The fraction of time spent in each state (set by binding/unbinding rates between filament and cytoplasm) acts as an order parameter for the efficiency of the long-distance transfer of materials. In the case that the motors are not allowed to diffuse in the surrounding medium after detachment from filaments, they just halt motion on the cytoskeleton. We address the influence of such waiting times on the transport properties.

P.30 Protein films at tailored interfaces: Towards controlling adsorption and arrangement

Hendrik Hähl

Experimental Physics, Saarland University, Saarbrücken, Germany

In a biological environment, proteins are usually the first molecules covering new interfaces. They form a conditioning film for further biofilm development. We study the development of the protein film from single protein adsorption to the final film. Thereby, we aim at controlling the adsorption behavior to interfaces by characterizing and controlling the interactions present between all involved partners, i.e. proteins, solution and substrate's surface.

In case of the solid/liquid interface, our studies revealed that for protein film formation, not only the actual surface characteristics (charge, surface energy,..) are important, but also the exact knowledge of the subsurface properties [1,2]: Long-ranged van der Waals forces, which are determined by the electrodynamic response of the involved materials, impact protein density and surface mobility. The experiments have been performed with BSA, lysozyme, and amylase on stratified substrates (Si/SiO_2), where the separation of surface and subsurface effects was possible. All three proteins revealed a similar behavior in protein density and film thickness upon changes in short- and long-range forces [1].

For characterizing protein/protein interactions, we have chosen amphiphilic proteins (hydrophobins) that exhibit specific ordering phenomena at interfaces [3]. Using wild types and engineered variants of hydrophobins, their form, charge distribution, and polar group arrangement could be varied. With this set of proteins, we study the adsorption kinetics and the protein film structure by ellipsometry, X-ray reflectometry and atomic force microscopy, all *in situ*. Peculiar behavior of the hydrophobins is that after a constant adsorption rate to the air/liquid interface, an abrupt transition to a saturated layer is observed [4]. We find a leading influence of steric interactions on the lateral arrangement of the proteins.

[1] H. Hähl, F. Evers, S. Grandthyll, M. Paulus, C. Sternemann, P. Loskill, M. Lessel, A. K. Hüsecken, T. Brenner, M. Tolan and K. Jacobs, *Langmuir* **28** (2012) 7747.

[2] P. Loskill, H. Hähl, T. Faidt, S. Grandthyll, F. Müller, and K. Jacobs, *Adv. Coll. Interf. Sci.* **107** (2012) 179182.

[3] S. Varjonen, P. Laaksonen, A. Paananen, H. Valo, H. Hähl, T. Laaksonen and M. Ben Linder, *Soft Matter* **7** (2011) 2402.

[4] (manuscript in preparation).

P. 31 Integration of genetic and epigenetic data into cellular networks identifies major determinants associated with breast invasive carcinoma

Mohamed Hamed

Centre for Bioinformatics (CBI), Saarland University Saarbrücken, Germany

Breast cancer is a genetically heterogeneous type of cancer that belongs to the most prevalent and death causing types. Treatment and prognosis of breast cancer relies largely on a correct classification and identification of the major determinants driving the tumorigenesis process. In the light of the availability of tumor genomic data from different sources and experiments, new integrative approaches are needed to boost the probability of identifying the associated genetic key drivers. We present here an integrative network based approach that is able to associate regulatory network interactions with the development of breast carcinoma by integrating information from gene expression, DNA methylation, miRNA expression, and somatic mutation datasets. Thereby, we suggest new genetic key drivers/determinants of breast cancer that could be further investigated in the wet lab as potential drug targets. This integrative network based approach can be applied in a similar fashion to other cancer types or complex diseases or for studying cellular differentiation and reprogramming.

P.32 The calcium dependence of target cell killing by primary human immune cells

Xiao Zhou, Yan Zhou, Rouven Schoppmey, Kim Friedmann, Cora Stephan, Sebastian Mang, Christian S. Backes, Carmen Mangerich, Hélène Lyrmann, Arne Knörck, Shruthi S. Bhat, Adrian Angenendt, Annette Lis, Bin Qu, Carsten Kummerow, Eva C. Schwarz, Markus Hoth

Department of Biophysics, School of Medicine, Saarland University, Homburg, Germany

Natural killer (NK) cells and cytotoxic T lymphocytes (CTL) form an immune synapse (IS) with their cognate target cells to kill them through the synaptic release of lytic granules (LG) containing perforin and granzymes or through Fas/FasL interaction. Quantifying target cell lysis over time, we observed two kinetically different killing phases in CTL and NK cells (Kummerow et al., 2014). Using siRNA against perforin, we found that the first and faster phase can be almost completely inhibited by perforin depletion. Consistent with these results, the first phase could also be blocked by very low concentrations of concanamycin A (10 nM), which inhibits perforin-based cytotoxicity due to its degradation by a pH increase in LG, but leaves Fas/FasL dependent cytotoxicity intact, which we found to be responsible for the second slower killing phase by analyzing Fas-deficient target cells. We conclude that the fast killing phase is perforin-dependent.

IS formation induces the activation of store-operated Ca^{2+} entry in NK cells and CTL through STIM-activated ORAI channels. Ca^{2+} entry through ORAI channels modulates perforin-dependent target cell killing but the Ca^{2+} dependence of target cell killing and the molecular mechanisms how Ca^{2+} controls NK cell and CTL cytotoxicity are not known. Therefore it is important to characterize and quantify the Ca^{2+} dependence of target-cell killing. The Ca^{2+} dependence of CTL and NK-mediated target cell killing was tested by changing the magnitude of Ca^{2+} influx through Orai channels applying different extracellular Ca^{2+} concentrations. We found that the fast, perforin-dependent killing phase is highly Ca^{2+} dependent with an optimal killing efficiency

between 0.16 and 0.65 mM external Ca^{2+} for CTL. These values are below free Ca^{2+} levels in the blood (ca. 1.2 mM) but in the range of external Ca^{2+} gradients reported in different tissues including skin. Similar experiments with NK cells revealed that their Ca^{2+} optimum to induce cell death is shifted to higher values compared to CTL.
One possibility to explain these findings is that the exocytosis of LG containing perforin and/or granzymes is Ca^{2+} dependent and shows a non-linear Ca^{2+} dependence in CTL and NK cells. We therefore tested exocytosis of single LG containing perforin-mCherry and/or granzyme B-mCherry with total internal reflection fluorescence microscopy at a model immune synapse by coating stimulatory anti-CD3/anti-CD28/anti-LFA-1 antibodies on a glass coverslip. Preliminary data show that the exocytosis probability of perforin- and granzyme B-containing LG is indeed Ca^{2+} dependent in the physiological range. The data are consistent with a bell-shaped Ca^{2+} dependence. The Ca^{2+} dependence of perforin and granzyme B release was also tested with an independent approach (ELISA assay), the results essentially confirmed the TIRF findings. We hypothesize that the bell-shaped Ca^{2+} dependence of LG release contributes to the Ca^{2+} dependence of perforin/granzyme-dependent target cell killing by CTL and NK.

P.33 Numerical analysis of hydrophobin adsorption at interfaces

Philipp Hudalla, Hendrik Hähl, Ludger Santen, Karin Jacobs

Theoretical Physics, Saarland University, Saarbrücken, Germany

Hydrophobins are strongly amphiphilic proteins which are known for their high surface activity and exhibit very linear and slower-than-langmuir adsorption kinetics at the air-water-interface. These uncommon kinetics however are sensible to manipulations of either the protein itself or the ion strength in the solution, suggesting an influence of crystallization effects and electrostatic interactions.
In order to explain the underlying mechanism behind this behaviour a bilayer lattice gas model is developed, describing a reservoir of mobile particles next to the interface. We show that for sufficient reservoir

mobility several features of the hydrophobin adsorption can be recreated in this model by effectively adjusting the local concentration next to the interface with this preadsorption mechanism.
Taking protein-protein interactions into account allows us to increase the geometric avail- ability of unoccupied sites at the interface and thereby achieving better agreement with the observed constant adsorption rate. We identify the relevant features of interaction potentials which could lead to the desired dynamics by such a "guiding effect".

P.34 Mechanical modeling of collective sheet migration

Mandar M. Inamdar

Indian Institute of Technology Bombay, Mumbai, India

In multicellular organisms the cells are typically organized collectively in tissues and are greatly constrained in their movements due to inter-cellular attachments. As a result, single cell motility studies are insufficient to understand the collective cell migration behavior. Since the phenomenon of collective cell migration arises in many biological processes such as morphogenesis, wound healing, as well as cancer growth, it is a topic of active current research. A particular form of collective cell migration called *sheet motility*, which, as the name suggests, involves cells moving together in a flat monolayer, is widely studied with *in-vitro* wound healing assays. In these experiments, a confined confluent colony of epithelial cells is exposed to free boundaries in order to initiate collective sheet migration. These experiments demonstrate that subsequent to exposure to free boundaries, the cells exhibit complex velocity patterns and exert intricate tractions on the underlying substrate. In addition, it was further demonstrated that migrating sheets exhibit periodic stripes of mechanical waves originating from the sheet boundaries that may have a role to play in specifying complex differentiating patterns during embryonic gastrulation. To address these complex experimental observations, we have developed a simple physical model of a self-

propelled, heterogeneous elastic membrane sliding over a viscous substrate. Our model can qualitatively explain a few experimentally observed facts: i) the swirling pattern of velocities in the interior of the tissue, ii) rigid body rotation of confined tissues, and iii) finger-like roughening of the tissue boundaries. Work is currently underway to also understand the traction patterns and stress-velocity correlations observed experimentally.

P.35 Dynamics of diffusing particles interacting with directionally moving particles on a polar filament

Denis Johann, Debajit Goswami, Karsten Kruse

Theoretical Physics, Saarland University, Saarbrücken, Germany

During cell division, pairing of sister chromosomes and their segregation is driven by the mitotic spindle, which is a stable bipolar structure consisting of overlapping microtubules, motors and other associated proteins. However, how the interplay between these proteins and microtubules render stability to the spindle structure is still unknown. Passive mobile cross-linkers called Ase1 have been found to dynamically adapt the dynamics of microtubule sliding induced by molecular motors to the length of the microtubules' overlap region [1].

Here, we study the effect of Ase1 on molecular motors in the presence of steric interactions in terms of a stochastic lattice model. We find that the Ase1 accumulate towards the end of the lattice in the direction of motor hopping and characterize their distribution through a mean-field theory.

[1] Braun et al., Nat. Cell. Biol. **13**, 1259 (2011).

P.36 *Staphylococcus aureus*: cell wall elasticity and turgor pressure

Philipp Jung[1], Henrik Peisker[1], Nicolas Thewes[2], Peter M Loskill [2,4], Pedro M Pereira[3], Mariana G Pinho[3], Markus Bischoff[1], Karin Jacobs[2] and Mathias Herrmann[1]

[1]*Institute of Medical Microbiology and Hygiene, Saarland University, Homburg, Germany*

[2]*Experimental Physics, Saarland University, Saarbrücken, Germany*

[3]*Instituto de Tecnologia Quımica e Biologica, Universidade Nova de Lisboa, Lisbon, Portugal*

[4]*Healy Laboratory, Department of Bioengineering, University of California, Berkeley CA, USA*

The *S. aureus* cell wall is of crucial importance for the bacterial survival and environmental interaction. It functions as a protection against chemical and mechanical stress, plays an important role in pathogenicity and fulfills a variety of physiological functions. Major components of the *S. aureus* cell wall are peptidoglycan strains, cross-linked via oligopeptid bridges. The crosslinking is catalyzed by enzymes with transpeptidase and transglycosidase active enzymes, termed penicillin binding proteins (Pbp). Additionally, the cell wall is interspersed with cell wall and cell membrane anchored proteins, protein pores and lipo/wall teichoic acids. Another important component of the cell wall plasticity is cell turgor pressure, which is formed by the passage of water through the cell wall due to osmotic active substances inside the bacterial cell. By pushing the cell membrane against the inner cell wall, the turgor pressure contributes substantially to the cell shape and integrity of the cell wall.
Here, we aimed to study the impact of the cell wall crosslinking and wall teichoic acids on the cell wall elasticity and the interplay between cell wall elasticity and cell turgor pressure by Atomic Force elasticity mapping of living *S. aureus* cells. In this context we investigated *S. aureus* wild type strains and isogenic deletion mutants lacking *pbp4*, encoding a transpeptidase involved in the secondary crosslinking of the cell wall, and *tagO*, encoding for the first enzymatic step of the wall teichoic acid biosynthesis. Further, the influence of proteins on

elasticity of the bacterial envelope was determined by performing nanomechanical mapping on purified peptidoglycan.
The *pbp4* deletion-induced decrease in cell wall crosslinking resulted in a significant increase of the cell wall elasticity, suggesting that the cell wall crosslinking markedly contributes to the cell wall stiffness. Experiments on purified peptidoglycan showed a negative effect on elasticity with twofold increase in Young's modulus most probably due to the lack of proteins in the peptidoglycan matrix. However, no clear correlation between the absence or presence of wall teichoic acids and cell wall elasticity was found, contrary to the current understanding of the function of wall teichoic acids in this process.
Interestingly, by increasing the contact force of the AFM probe from 1 nN to 5 nN during elasticity mapping, elasticity information independent of the cell wall properties was obtained, which responded to changes of the osmotic environment, suggesting that cell turgor pressure properties can be determined by this type of contact force alteration.

P.37 Conditional and selective activation of the Gq pathway in the heart and its impact on cardiac function in vivo

Elisabeth Kaiser, Jürgen Wess, Lars Kaestner, Peter Lipp

Molecular Cell Biology, Saarland University, Homburg, Germany

Questions/Aims: The specific impact of Gq signaling on the mammalian heart, especially on the conduction system, is largely unidentified. We therefor aim to examine how Gq activation alters cardiac physiology by inducing Gq receptor activation specifically in cardiomyocytes.
Methods: We investigate a novel transgenic mouseline, expressing the designerreceptor r/hM3Dq(a so-called DREADD) controlled by the MCK promoter. In these animals the Gq pathway can be selectively activated in striated muscle cells including cardiomyocytes through injection of their highly specific, synthetic and otherwise inert agonist CNO (Clozapine-N-Oxide).

Results: Cardiac function was studied in-vivo by telemetric devices (DatasciTM). We found that r/hM3Dq+/- mice develop immediate cardiac arrhythmias in a CNO dose-dependent manner. This development was simultaneous to an increase in the mean aortic pressure. CNO application to isolated cardiomyocytes resulted in augmented electrically induced Ca2+ transients.
Conclusions: Expression of r/hM3Dq leads to massive alterations in cardiac pacemaking, impulse propagation and Ca^{2+} handling. This approach offers therefor a powerful method to investigate the role of Gq signaling on the cardiac conduction system and (patho-) physiologic heart performance, which is especially eligible under in vivo conditions.

P.38 Mechanics of the nucleus dictated by the cytoskeleton

Susanne Karsch and Florian Rehfeldt

3rd Institute of Physics – Biophysics, Georg-August University, Göttingen, Germany

The nucleus, confined by the nuclear envelope, consisting of two lipid bilayer membranes and a protein network made up by e.g. lamins, creates a specific microenvironment for the genome. It has multiple crosslink proteins which connect the nucleus to the cellular cytoskeleton and further via focal adhesions to the extra-cellular environment. Thus, the nucleus is an integral part of the mechano-sensing machinery of the cell and has the ability to respond to mechanical signals from the cell`s surrounding. However, the exact mechanisms how shape, mechanics and structural organization respond to the outside is not fully understood. Elucidating these characteristics is of high importance as it directly impacts gene expression and could be related to certain diseases.
Here, we analyze the mechanical properties of the nucleus focusing on the influence of the cytoskeleton. Our approach of combining measurements of isolated nuclei and nuclei in cells gives insights into this interplay. Imaging techniques together with atomic force microscopy allow us to examine not only morphology and structure of

the nucleus but we can also assess the mechanics. Using these techniques we quantify the mechanical properties of the nucleus and show the significant impact of the cytoskeleton.

P.39 Environmental control of stochastic cargo transport by teams of molecular motors

Sarah Klein[1,2], Cécile Appert-Rolland[2], Ludger Santen[1]

[1] *Theoretical Physics, Saarland University, Saarbrücken, Germany*

[2] *Laboratoire de Physique Théorique, CNRS, Université Paris-Sud, Orsay, France*

Many different types of cellular cargos are transported bidirectionally along microtubules by teams of molecular motors. The motion of this cargo-motors system has been experimentally characterized in vivo as processive with rather persistent directionality. By means of an effective theoretical approach, introduced by Lipowsky et al. [1], it has been argued that the dynamics of these object are the result of a tug-of-war between different kinds of motors. This picture has been questioned in a recent article by Kunwar et al. [2], who considered the coupling between motorand cargo in more detail.
Based on this framework we introduce a model regarding single motor positions. We show that bias can be simply controlled or even reversed in a counter-intuitive manner via a change in the external force exerted on the cargo or a variation of the ATP binding rate to motors. Furthermore, the superdiffusive behavior found at short time scales indicates the emergence of motor cooperation induced by cargo-mediated coupling.

[1] M. J. I. Müller, S. Klumpp, and R. Lipowsky. Tug-of-war as a cooperative mechanism for bidirectional cargo transport by molecular motors. *Proceedings of the National Academy of Sciences*, 105(12):4609–4614, Mar. 2008.

[2] A. Kunwar, S. K. Tripathy, J. Xu, M. K. Mattson, P. Anand, R. Sigua, M. Vershinin, R. J. McKenney, C. C. Yu, A. Mogilner, and S. P. Gross. Mechanical stochastic tug-of-war models cannot explain bidirectional lipid-droplet transport. *Proceedings of the National Academy of Sciences*, 08(47):18960–18965, 2011.

P.40 Ca^{2+} dependence of $CD8^+$ T cell proliferation and subset development

Knörck A., Friedmann K., Hoth M., Schwarz E.C.

Department of Biophysics, School of Medicine, Saarland University, Homburg, Germany

Ca^{2+} signals control the transcriptional activity of T cells thus regulating the activation, development and proliferation of T cells. Cytotoxic T lymphocytes (CTL) eliminate target cells by the release of lytic granules which is known to be Ca^{2+}-dependent. As Ca^{2+} signals control also proliferation, we analyzed proliferation of $CD8^+$ T cells following stimulation with anti-CD3/anti-CD28 antibody coated beads. Different [Ca^{2+}] within the medium were adjusted either by the addition of EGTA or CaCl2. The analysis of six different donors showed that the proliferation is strongly dependent on extracellular [Ca^{2+}] over four days with similar Ca^{2+} dependencies in the donors although individual proliferation rates differed. Proliferation was totally wiped out with the addition of 1.5 mM EGTA whereas the addition of Ca^{2+} up to 4 mM still enhanced proliferation rates. To determine the free [Ca^{2+}] in AIMV medium (containing 10% FCS) under different conditions (EGTA or Ca^{2+} added), we used three different methods: measurements with a Ca^{2+}-sensitive electrode, the Ca^{2+}-sensitive fluorescent dye Mag-Fura-2 and for higher [Ca^{2+}] (not more than 0.4 mM EGTA added) with a blood gas analyzer. To analyze the $CD8^+$ T cell subset development after in vitro stimulation we used different surface markers for cell characterization. Flow cytometry was carried out using anti-CD3, -CD8, -CD27, -CD28, -CD197, -CD45RA and -perforin labeled antibodies to compare different stimulation methods either by anti-CD3/anti-CD28 coated beads or *Staphylococcus aureus* enterotoxin A (SEA). The stimulation of $CD8^+$ T cells by SEA generated a large pool of the effector-memory subpopulation. It will be interesting to combine the Ca^{2+} dependent proliferation data with the subpopulation development to test, if Ca^{2+} shifts the relative subpopulation proportions.

P.41 Calcium and ROS: from detection to function

Maik Konrad[1], Xin Zhang[1], Anjana Rao[2], Xin Hui[3], Peter Lipp[3], Vsevolod V. Belousov[4], Barbara A. Niemeyer[1], Ivan Bogeski[1]

[1]*Department of Biophysics, Saarland University, Homburg, Germany*

[2]*La Jolla Institute for Allergy and Immunology, La Jolla, California CA, USA*

[3]*Institute for Molecular Cell Biology, Saarland University, Homburg, Germany*

[4]*Shemyakin and Ovchinnikov Institute of bioorganic chemistry, Moscow, Russia*

T cell activation, differentiation and proliferation, are processes which critically depend on store-operated Ca^{2+}-entry (SOCE) mediated by stromal interaction molecules (STIM) and the Orai ion channels. The Orai dependent activation of T cells also leads to local production of intracellular reactive oxygen species (ROS), the physiological function of which is not well understood. In a recent study we revealed that oxidation of critical cysteine residue of Orai molecules will lead to inhibition of SOCE and hinder T cell activation. The translocation of NFAT, an important factor needed for successful T cell activation, is calcium dependent and requires a functional and long lasting activity of STIM-Orai mediated channel activity, impaired NFAT translocation may thus be due to an impaired calcium influx or homeostasis in T cells. In collaboration with A. Rao who has conducted a large siRNA based screen for defective translocation of NFAT to the nucleus, identified 6 redox active candidates which we set out to identify as possible regulators of SOCE. In addition, genetically encoded fluorescent sensors developed during the past decade provided with novel tools to investigate intracellular calcium signaling and ROS generation between different compartments. The simultaneous use of different sensors may grant us the opportunity for real time detection and delineation of these two concomitant events in T cell activation, proliferation, apoptosis and other processes. In cooperation with different groups we are trying to develop new and improve existing mitochondrial calcium sensors. We have also developed and tested novel redox-sensitive

fluorescent sensor HyPer and HyPer Red to measure mitochondrial ROS production in established cell lines including T cells and melanoma cells.
By utilizing, developing and improving different techniques for our needs we hope to unravel the interplay between ROS and calcium signals for a better understanding of the human immune system.

P.42 A technique to distinguish two modes of immune cell killing on single cell level

Christian Backes, Sebastian Mang, Rouven Schoppmeyer, Gertrud Schwär, Cora Stephan, Markus Hoth, Carsten Kummerow

Department of Biophysics, School of Medicine, Saarland University, Homburg, Germany

Death of cells in the body is involved in many diseases or injuries, but also occurs during development under physiological conditions. The killer cells of the immune system like cytotoxic T lymphocytes (CTL) or natural killer (NK) cells can eliminate malignant cancer cells or virus-infected cells by inducing cell death in their targets. Cell death can occur as a highly organized process during apoptosis or by plasma membrane disruption during necrosis. We have generated cell lines expressing a genetically-encoded sensor to quantify both necrosis and apoptosis in single living target cells by time-lapse fluorescent microscopy. We have observed that NK cells induce both types of cell death in a clonal population of target cells. Interestingly, individual NK cells can switch from necrosis to apoptosis during serial target cell killing. Furthermore, we have found that the induction of these two types of cell death is preceded by different calcium signals in the killer cell. Necrosis is associated with high and transient signals and apoptosis by lower and more sustained signals. We postulate that Ca^{2+} signals may regulate the mode of killing and that the relative contribution of apoptosis and necrosis is important in regulating the immune response towards cancer and infection.

We have also established a high-content protocol for this assay on an automated microscope and an analysis in a three-dimensional collagen matrix using light sheet fluorescence microscopy. This will enable us to use the assay for screening purposes and under conditions as physiological as possible.

P.43 Quantitative analysis of calcium dependent migration in human killer cells

Hélène Lyrmann[1], Marc Neef[2], Rouven Schoppmeyer[1], Christian Backes[1], Eva C. Schwarz[1], Bin Qu[1], Markus Hoth[1], Franziska Lautenschläger[3], Karsten Kruse[2] and Carsten Kummerow[1]

[1]*Department of Biophysics, School of Medicine, Saarland University, Homburg, Germany*

[2]*Theoretical Physics, Saarland University, Saarbrücken, Germany*

[3]*Experimental Physics, Saarland University, Saarbrücken, Germany*

The killer cells of the immune system, such as cytotoxic T lymphocytes (CTL) and natural killer cells (NK cells) search the body for virus-infected and cancerous cells. After recognition of a target cell, the killer cell releases lytic granules to kill the target. To optimize the immune function, the migration machinery of the killer cells has to be tuned to maximize the probability of target cell encounter.
We studied the calcium dependence of NK and CTL migration by time-lapse microscopy on a flat substrate (2D). Our analysis revealed that the cells alternate between a mobile and a stationary state, where the probability of each state depends on the external calcium concentration. On a short time scale (seconds/minutes), the killer cells show directional persistence, whereas migration on a longer time scale (hours) is random. This mode of migration can be modelled as an intermittent, persistent random walk. Simulations with this model reveal that, depending on the persistence time, a search time optimum can be reached in a given space for certain boundary conditions. We are

currently investigating the behavior of migrating killer cells upon contact with boundaries in 2D. Furthermore, we have started to study their migration in channels (1D) and in a collagen matrix (3D). We believe that modelling killer cell migration and search behavior can help us modify and optimize the immune response in health and disease.

P.44 Markov models for the (in)stable copying of DNA-methylation patterns in mammalian cells

Pascal Giehr[1], Charalampos Kyriakopoulos[2], David Spieler[2], Jörn Walter[1], Verena Wolf[2]

[1]Institute of Epigenetics, and [2]Computer Science Department, Saarland University, Saarbrücken, Germany

DNA methylation is an epigenetic modification associated with the regulation of gene function. Gene specific DNA-methylation patterns change over development and in specific cell types. DNA methyltransferases (Dnmts) in cooperation with other enzymes are responsible for the setting and maintenance of methyl groups at CpG-dinucleotides during cell division. The control of DNA methylation is highly dynamic involving regulated and random processes. We have generated a comprehensive model to describe the dynamics of methylation observed in dividing embryonic stem cells. The recent discovery of oxidative forms of DNA-methylation challenges some of these concepts. We have adapted our technology to also measure some of the oxidative forms. Such com- parative data allow deeper insights in the evolution of methylation patters in dividing and non-dividing cells. We propose a (hidden) Markov model to describe the dynamics of DNA methylation including its main oxidative form: hydroxymethylcytosine. Building on our previous models we are able to explain and interpret DNA methylation patterns measured by high resolution sequencing using the oxidative-hairpin-bisulfite technology (ox-hBis).

P.45 Generation of oscillating gene regulatory network modules

B. Lannoo , M. van Dorp , Q. Thommen , B. Pfeuty , M. Lefranc , E. Carlon

Institute for Theoretical Physics, Katholieke Universiteit Leuven, Leuven, Belgium

As a first step towards understanding the complex behavior of Gene regulatory networks (GRN), a lot of effort has been devoted in studying the dynamics of smaller modules in the past decades[2]. Here we focus on modules in which a target protein oscillates in time. Oscillatory behavior is found in many processes in the cell. For instance the circadian rhythms are driven by genetic modules [2], but there are also oscillations which are sub circadian like those in NF-kB, p53, Wnt.
For the production of these modules we used an improved version of the evolutionary algorithm proposed by François and Hakim [1]. The most crucial part of this algorithm is the design of a score function used to define which elements are considered the fittest. Here we show this score function, the simplest modules capable of showing oscillatory behavior [3] and the characteristics of the heterodimer autorepression loop module, which shows some interesting characteristics not yet discussed in the literature.

[1] Paul François and Vincent Hakim, *PNAS*, 101(2):580-585 2004.

[2] LH Hartwell, JJ Hopfield, S Leibler, and Murray AW. *Nature*, c47-c52, 1999.

[3] M. van Dorp, B. Lannoo, and E. Carlon. *Phys. Rev. E* 88:012722, Jul 2013.

P.46 Brownian dynamics simulations of the molecular encounter at the *E. coli pap* promoter

Po-Hsien Lee, Volkhard Helms

Center for Bioinformatics, Saarland University, Saarbrücken, Germany

The pyelonephritis-associated pili (*pap*) operon is a well-studied model for the regulation of a cluster of genes. The *pap* genes code for protein components of pili in *E. coli*. The three main proteins involved in regulating transcription of the *pap* operon are the leucine-responsive regulatory protein (Lrp), deoxyadenosine methylase (Dam) and PapI. Lrp proteins are supposed to form tetramers and attach to the DNA molecule at different binding sites to switch on/off the transcription of the *pap* gene. The binding affinity between Lrp and DNA is affected by Dam methylation of two GATC sequence stretches in the promoter region and also by the presence of PapI proteins. In this study, we represented Lrp proteins and the DNA molecule as vdW beads linked by harmonic springs. Brownian dynamics simulations are performed by employing binding affinities of various strengths for the protein-protein and protein-DNA interactions. Our results show that different protein-DNA oligomers were formed depending on the strength of the pairwise binding affinities. In the next step of this project, PapI will be taken into consideration to make our model system more realistic.

P.47 Visualizing immediate effects of neurosteroids DHEA and 7β-OH-DHEA on neurons by calcium imaging

Aline Löhfelm and Uli Müller

Biosciences Zoology/Physiology-Neurobiology ZHMB (Center of Human- and Molecular Biology), Saarland University, Saarbrücken, Germany

Neuroactive steroid hormones or neurosteroids with neuroprotective effects became more and more interesting in the last years due to their implication in aging and age- related neurodegenerative diseases as Alzheimer and Parkinson. The term neurosteroid was formed for neuroactive compounds produced *de novo* in the brain and for circulating steroids metabolized in the CNS to neuroactive forms. Although it is known that neurosteroids modulate ion-gated neurotransmitter receptors as GABAA or NMDA receptors, acetylcholine receptors were not yet studied intensively regarding neuroprotection. Using optical imaging this work aims to characterize immediate effects of the neurosteroids DHEA and 7β-OH-DHEA on calcium influx triggered by acetylcholin receptors. In this respect the effects of the neurosteroids on different subunit compositions of ACh-receptors and their expression in different types of neurons will be of special interest. Thus, several cell cultures are incubated with various inhibitory drugs to distinguish between sensitive and insensitive receptor types.

P.48 Exact and numerical results for a simple microtubule length regulation model

Chikashi Arita, Alexander Lück, and Ludger Santen

Theoretical Physics, Saarland University, Saarbrücken, Germany

The spatial organization and mechanical properties of cells depend strongly on the structure of the cytoskeleton. The cytoskeleton is a network of active biopolymers, which can undergo considerable morphological changes, for example during cell division. Therefore it is of great importance for the functioning of the cell to control these dynamic processes. One example of such a control mechanism is microtubule (MT) length regulation. MT length regulation can be carried out by kinesin motor proteins moving toward the growing MT end.

Recently stochastic models for the MT length regulation have been introduced [1,2], which are extended versions of the totally asymmetric simple exclusion process. These models take both the variable length of the filament and the influence of the motor proteins into account. Here we apply a matrix productansatz to the simplest case of the model introduced by Melbinger et al [2]. We obtain exact analytic results in the stationary state. In addition a rich phase diagram can be found by using Monte-Carlo simulations in the non-stationary state.

[1] D. Johann, C. Erlenkämper, and K. Kruse, Phys. Rev. Lett., 258103 (2012).

[2] A. Melbinger, L. Reese, and E.Frey, Phys. Rev. Lett., 258104 (2012).

P.49 A novel STIM2 splice variant functions as a break for STIM mediated activation of Orai calcium channels

Anna-Maria Miederer, Dalia Alansary, Gertrud Schwär, Martin Jung, Barbara A. Niemeyer

Department of Biophysics, Saarland University, Homburg, Germany

Cell homeostasis and Ca^{2+} signaling depend on a tight regulation of the intracellular Ca^{2+} concentration. Alterations in basal Ca^{2+} can lead to various diseases and likely contribute to development of abnormal growth. Different regulators such as calmodulin and Ca2+ pumps limit cytosolic [Ca^{2+}] and their down-regulation by siRNA lead to an increased basal [Ca^{2+}]. Another important regulator is the stromal interaction molecule 2 (STIM2) that shows a reduction in basal [Ca^{2+}] following knock down. The two known isoforms of STIM, STIM1 and STIM2, are ER resident membrane proteins which sense the Ca^{2+} content of the ER via their luminal EF-hands. After partial or complete store depletion STIM-proteins multimerize and trigger store-operated calcium entry (SOCE) by directly gating Orai channels localized at the plasma membrane. Here, we report the identification and characterization of a novel STIM2 splice variant, named STIM2.1, which differs in a single additional exon consisting of only 8 amino acids located within the STIM2 channel activating domain (CAD). We show the novel variant STIM2.1 is present in a variety of primary cells and cell lines although its relative expression varies in regard to the known variant (STIM2.2) and depends on the activation state and cell type. In contrast to STIM2.2, STIM2.1 is unable to gate Orai channels. Coexpression of STIM2.1 together with STIM1 displays reduced SOCE when compared to STIM2.2 coexpression. Splice variant specific knockdown of STIM2.1 in naïve human $CD4^+$ T cells increases SOCE whereas specific down-regulation of STIM2.2 decreases basal calcium as well as SOCE, suggesting that STIM2.1 acts as a negative regulator of STIM mediated SOCE. Biochemical experiments are being conducted to delineate the functional defects of STIM2.1.

P.50 DNA hybridization and kinetics of hairpin-loop molecules

Mina Mohammadi, Björn Ackermann, Albrecht Ott

Biologische Experimentalphysik, Saarland University, Saarbrücken, Germany

In the cell, molecular information processing is based on molecular recognition and binding. Although DNA hybridization is sometimes understood as 'lock and key'. It is not clear how two molecules can identify each other. We find that there are many possibilities of different single strands of DNA at a given length that can bind to a given surface bound probe in thermal equilibrium. In other words, many keys can coexist for one lock. There are some groups of sequences, which do not bind to a probe like the ones with runs of guanine bases or self-complementary sequences. At the same time we look at the behavior of corresponding DNA hairpin-loop molecules. We investigate their thermal fluctuation by using a combination of fluorescence energy transfer and fluorescence correlation spectroscopy. We measure the rate of opening and closing for different sequences with different stem or loop length. In future work we will look at the behavior of DNA hairpin-loop molecule in the presence of competitive targets to see how they identify their complementary probe. This is a first approach towards understanding how molecular recognition works in the crowded and competitive environment of the cell.

P.51 Identification of gene regulatory networks governing cellular identity

Maryam Nazarieh, Volkhard Helms

Center for Bioinformatics, Saarland University, Saarbrücken, Germany

Identification of gene regulatory networks governing cellular identity is one of the main problems for understanding the mechanism underlying

cellular differentiation and reprogramming. In this work, we transfer this problem to an optimization problem, namely determining a Minimum Connected Dominating Set for directed graphs. This is motivated by the biological observation that the pluripotency network in embryonic stem cells is maintained by a few transcription factors which share many target genes. Finding a subset of genes out of 2^n subsets of genes takes exponential time, but approximation algorithms have been developed which find a close to optimal solution in polynomial time with a constant approximation factor. Experimental results on time-series gene expression data during a cell cycle show that this method can identify top regulators which govern the cell cycle in the model organisms Ecoli and Yeast.

P.52 Roll instabilities of an active polar fluid in the planar Taylor-Couette geometry

Marc Neef and Karsten Kruse

Theoretical Physics, Saarland University, Saarbrücken, Germany

Biological matter, e.g. cell tissues or the cytoskeleton, can flow spontaneously due to their active microscopic properties. A general theory for active materials based on fundamental physical principles like conservation laws and geometrical invariances has been developed by Kruse et al. in [1].
We study the dynamics of active polar fluids in a Taylor-Couette geometry where the fluid is confined between two coaxial cylinders. We find that, depending on the system's activity, complex flow patterns – stationary as well as rotating – can arise spontaneously.

[1] K Kruse et al., Eur. Phys. J. E 16, 5 (2005).

[2] S Fürthauer et al., New J. Phys. 14, 023001 (2012).

P.53 ATP concentration regulates cellular processes in time and space

Jasmine A. Nirody[1], and Padmini Rangamani[2]

[1]Biophysics Graduate Group and [2]Department of Molecular and Cellular Biology, University of California, Berkeley CA, USA

Adenosine 5'-triphosphate (ATP) is the primary "energy currency" of most known living organisms, and is a crucial participant in the majority of enzymatic reactions. The concentration of ATP is assumed to be constant in standard enzyme kinetics because it is assumed to be always present in large excess. This allows enzyme-mediated processes to be modeled as single- substrate reactions, greatly simplifying the associated mathematical analyses. However, this assumption fails to hold when ATP levels are comparable to substrate levels. Such situations may arise via mitochondrial dysfunction [1] or spatial inhomogeneity in ATP concentrations [2]. Indeed, the distribution of ATP in a cell has been strongly suggested to have a regulatory role in several cellular processes, including the excitation-contraction coupling in skeletal muscle [3] and actomyosin dynamics [2]. Here, we demonstrate the importance of ATP concentration on the dynamics of multi-enzyme reactions by explicit consideration of ATP in enzyme-substrate reactions. Preliminary results indicate the concentration of ATP plays in important role in determining both the time scale and equilibrium concentrations of the substrate and product. We apply our computational framework to study the dynamics of mitogen-activated protein (MAP) kinase cascade, which is involved in the regulation of a vast range of cellular activities [4]. In this study, we characterize the role of the spatial and temporal cellular energy landscape in governing this ubiquitous cellular process.

[1] Berg JM, Tymoczko JL, Stryer L. *Biochemistry*, W.H. Freeman, 2010.

[2] van Horssen R *et al.* Modulation of cell motility by spatial repositioning of enzymatic ATP/ADP exchange capacity. *J Biol Chem*, 284(1), 2009: 1620-1627.

[3] Dutka TL, Lamb GD. Effect of low cytoplasmic [ATP] on excitation-contraction coupling in fast-twitch muscle fibres of the rat. *J Physiol* 560(2), 2004: 451-468.

[4] Seger R, Krebs EG. The MAPK signaling cascade. *FASEB J.* 9(9), 1995: 726-735.

P.54 Properties of novel voltage sensitive dyes in adult ventricular myocytes

Katja Nitze[1], Qinghai Tian[1], Ping Yan[2], Leslie Loew[2], Peter Lipp[1], Lars Kaestner[1]

[1]*Institute for Molecular Cell Biology, Saarland University, Homburg, Germany*

[2]*R.D. Berlin Center for Cell Analysis and Modeling, University of Connecticut Health Center, Farmington CT, USA*

The gold standard for studying the electrical activity of excitable cells is the patch clamp technique either in voltage-clamp or current-clamp mode. The latter approach allows investigation of action potentials in excitable cells such as cardiac myocytes or neurones. It though requires an electrode to be inserted into the cells and to rupture a part of the cell membrane. In contrast, fluorescent dyes can help visualizing action potentials in a non---invasive manner. Fast voltage probes such as Di-8-Anepps integrate into cell membranes and sense changes in the transmembrane voltage, i.e. they change their spectral properties with changes in membrane potential.

We investigated the efficiency of novel dyes that were designed to emit light shifted more to longer wavelengths of the spectrum than dyes available previously using voltage clamp of isolated mouse ventricular myocytes in combination with spectrally resolved fluorescence photometry. Therefore we recorded both excitation and emission spectra upon defined changes in membrane voltage. Our analysis of these novel voltage sensors revealed interesting structure-function relationships, e.g. the linker length correlates with the membrane potential dependent shift of the emission spectra. Such optimized small molecule voltage probes will allow simultaneous ratiometric measurements of the membrane potential, the intracellular calcium

concentration and contraction in cardiac myocytes with increased signal-to-noise ratios.

P.55 Subharmonic oscillations of collective molecular motors

D. Oriola[1], H. Gadêlha[2,3], C. Blanch-Mercader[1], and J. Casademunt[1]

[1]*Departament d'Estructura i Constituents de la Matèria, Facultat de Física, Universitat de Barcelona, Barcelona, Spain*

[2]*Department of Applied Mathematics and Theoretical Physics, University of Cambridge, Cambridge*, United Kingdom

[3]*Wolfson Centre for Mathematical Biology, Mathematical Institute, University of Oxford, Oxford, United Kingdom*

The collective action of molecular motors can often exhibit instabilities leading to oscillatory behaviour. Some examples are the periodic beating of cilia and flagella, oscillatory regimes in muscles, or the spontaneous oscillations of auditory hair cells. Here we study a generic two-state model for an assembly of molecular motors described by means of a pair of integro-partial differential equations, which leads to oscillatory motion in the presence of an elastic load [1]. We reduce the system to a minimal set of three ordinary differential equations and we study in detail the effective model, which we show that captures the main nonlinear dynamics of the full system. In the high mobility regime and for strong elastic modulus, we report on the emergence of subharmonics in the power spectrum of the oscillations that could account for unexplained experimental observations in a minimal *in vitro* actomyosin system [2].

[1] Jülicher F. and Prost J., *Phys. Rev. Lett.*, 78, 4510 (1997).

[2] Plaçais P.-Y., Balland M., Guérin T., Joanny J.-F. and Martin P., *Phys. Rev. Lett.*, 103, 158102 (2009).

P.56 Dynamics of human blood platelets on elastic and patterned substrates

Aishwarya Paknikar, Rabea Sandmann, and Sarah Köster

Institute for X-Ray Physics, Georg-August-Universität Göttingen, Göttingen, Germany

During a bleeding event, platelets mediate blood clotting by rearranging their actin-myosin cytoskeleton to generate forces and contract. *In vivo*, platelets function in a variety of mechanical environments ranging from soft tissues to stiff and structured wound surfaces. The mechanical response of platelets to such different environments is elusive and hence we investigate the behavior of single platelets on flat, elastic (E_{sub} in physiological range of 1-100 kPa) polyacrylamide (PAA) gels and stiffer, microstructured polydimethylsiloxane (PDMS) substrates. In particular, we study the spreading dynamics, formation of cellular protrusions and contractile forces. We have observed that the final platelet spread area increases with increasing values of E_{sub} and the most pronounced sensitivity to stiffness lies between 0-40 kPa. Furthermore, spreading on flat substrates is faster than on structured ones. While spreading, the platelets on structured substrates adapt their membrane shape to the underlying topography [1]. The spread area over time shows a sigmoidal course and during spreading the platelets retract over the holes at their periphery and compensate for the area losses by greater spreading on the interspaces between the holes. This observation leads us to the conclusion that platelets can detect their spread area and by keeping it constant ensure a controlled distribution of cellular components. During spreading, the contractile forces of the platelets increase. The average total force of a single platelet on a soft PAA substrate, measured by traction force microscopy (TFM), is ~34 nN and myosin contributes majorly to the total force generation [2]. We use TFM to analyze the influence of varying PAA substrate elasticities on the platelet force fields. Our preliminary observations indicate that the total force of a platelet increases linearly before reaching a steady state. We further analyze the relationship between force curve saturation point and spread area curve saturation point of a single platelet on

varying PAA stiffnesses. Our experimental findings aim at building a mechanical model for platelet dynamics on a single-cell level.

[1] R. Sandmann, S. Schwarz G.Henriques, F. Rehfeldt. (2013). Micro-Topography influences Blood Platelet spreading. Soft Matter, 10, 2365-2371.

[2] S. Schwarz G.Henriques, R. Sandmann, A. Strate, and S. Köster. (2012). Force field evolution during human blood platelet activation. Journal of Cell Science, 125, 3914–3920.

P.57 Detecting protein complex subunits in whole eukaryotic cells in aqueous environment

Diana B. Peckys[1], Ulrike Korf[2] and Niels de Jonge[1]

[1]INM – Leibniz Institute for New Materials, Innovate Electron Microscopy, Saarbrücken, Germany

[2]German Cancer Research Center DKFZ, Division of Molecular Genome Analysis, Heidelberg, Germany

We have used scanning transmission electron microscopy (STEM) of cells in liquid [1,2], so-called Liquid STEM, to determine the stoichiometry of protein complexes directly on whole cells. Liquid STEM was used to study the epidermal growth factor receptor (EGFR), and the receptor tyrosine kinase ErbB2. The distribution of monomers, homodimers, and heterodimers of these receptors is of relevance for basic research and for the analysis of anti-cancer drug mechanisms. COS7 fibroblast cells and SKBR3 breast cancer cells were grown on silicon microchips with silicon nitride (SiN) membrane windows, and incubated with specific, small protein ligands conjugated to gold nanoparticles or to fluorescent quantum dots (QDs). Using correlative fluorescence microscopy, we confirmed the known phenomenon of heterogeneity in non-isogenic cancer cells. We found not only SKBR3 cells to display very diverse morphologies, but also large variations in EGFR and ErbB2 expression levels and their spatial distributions on the cell membrane. Fluorescence images were used to pre-select individual cells and cellular regions of interest for high-resolution electron microscopy in wet state, performed with an environmental scanning

electron microscope, equipped with a STEM detector. Since the Liquid STEM method avoids the extensive sample preparation common for electron microscopy of biological samples, intact and large areas of cell membrane could be imaged from statistically relevant numbers of cells. The achieved resolution allowed protein complexes to be studied at the level of individual subunits. Liquid STEM thus provided data from thousands of labels, which were analyzed by automated image software algorithms [3].

[1] de Jonge, N., Peckys, D.B., Kremers, G.J., Piston, D.W., Proc Natl Acad Sci 106, 2159-2164, 2009.

[2] de Jonge, N., Ross, F.M., Nature Nanotechnology 6, 695-704, 2011.

[3] Peckys, D.B., Baudoin, J.P., Eder, M., Werner, U., de Jonge, N., Scientific reports 3, 2626, 2013.

P.58 Interplay of channels, pumps and organelle location in calcium microdomain formation

M. Peglow[1], I. Maccari[1], B. Niemeyer[2], M. Hoth[2], H. Rieger[1]

[1]*Theoretical Physics, Saarland University, Saarbrücken, Germany*

[2]*Biophysics, Saarland University, Homburg, Germany*

To analyze the influence of Calcium (Ca) microdomains on the global cytosolic Ca concentration, we consider the polarization and activation of T-cells after the formation of an immunological synapse as a model system. For T-cell proliferation and activation, a high and robust Ca signal lasting from minutes up to hours is needed. This raises the intriguing question of how T-cells overcome all those mechanisms which normally remove an increased Ca level as fast as possible from the cytosol. With the help of a theoretical model [1] we predict that, after the formation of a local Ca influx pathway via STIM1 and Orai1, mitochondria relocation toward and accumulation of plasma membrane Ca-ATPase and sarcoplasmic/endoplasmic reticulum calcium ATPase pumps at the immunological synapse are sufficient to achieve a long lasting increased global Ca concentration.

[1] M. Peglow, B. Niemeyer, M. Hoth, H. Rieger: New J. Phys. 15, 055022 (2013)

P.59 The role of Orai3 in SOCE of prostate cancer cells

Tatiana Kilch[1], Sven Kappel[1], Volker Jung[2], Michael Stöckle[2], Ivan Bogeski[1] and Christine Peinelt[1]

[1]*Department of Biophysics, ZHMB, Saarland University, Homburg, Germany*

[2]*Clinics of Urology and Pediatric Urology, Saarland University, Homburg, Germany*

Reactive oxygen species (ROS) are elevated in prostatitis and prostate cancer and contribute to altered cancer hallmark functions such as enhanced epithelial cell proliferation and neoplasia. Even though ROS inducing substances and ROS scavengers have been investigated as therapeutics the outcome and benefit of such strategies remains largely unclear and vague. Therefore a better understanding of the underlying mechanisms and key players in ROS mediated signaling pathways is required for future therapeutic approaches. There is a clear role for Ca^{2+} in ROS mediated signaling and it is known that store-operated Ca^{2+} entry (SOCE) is impaired in prostate cancer.

Here we investigate the relationship between Orai subunit ratios Orai3/Orai1 and ROS induced inhibition of SOCE in androgen-sensitive and androgen-insensitive prostate cancer cell lines. Our data indicate a clear role for Orai3 in SOCE signaling and support the concept that indeed, the Orai3/Orai1 ratio correlates with redox sensitivity of SOCE.

P.60 The extracellular adherence protein of *Staphylococcus aureus* - new functions of a multifunctional adhesin

Henrik Peisker[1], Mona Saffarzadeh[2], Markus Bischoff[1], Mathias Herrmann[1], and Klaus Preissner[2]

[1]*Institute of Medical Microbiology and Hygiene, Saarland University, Homburg, Germany*

[2]*Department of Biochemistry, Medical School, Justus-Liebig-University, Giessen, Germany*

The *Staphylococcus aureus* extracellular adherence protein (Eap) is part of the SERAM (Secretable expanded repertoire adhesive molecules) termed protein family and suggested to play a significant and multifunctional part in the pathogenicity and wound infection in *S. aureus*. Eap has been shown to bind to a number of host cell matrix and plasma proteins and to facilitate internalization in eukaryotic cells by providing an anchoring function that may mediate subsequent bacterial uptake. Immunomodulatory and antiangiogenic functions have also been attributed to Eap as well as a delayed wound healing. Here we show data on two new functions of the adhesin Eap obtained by atomic force microscopy (AFM) and force sensitive nanomechanical mapping (PF-QNM). We conducted two experiments to determine new functions of Eap.

First, Eap incubated HaCat cells were analyzed regarding cell elasticity, cell volume, surface area and projected surface area as well as cytoskeleton orientation and integrity. AFM and fluorescence images show significant changes in cytoskeletal organization as well as increased elasticity and geometrical changes for Eap incubated cells when compared to the control. While the cell volume decreased, surface area and projected surface area as well as boundary length increased drastically resulting in spread cell geometry. We speculate the Eap induced increase in flexibility results in an increase of cell to surface contact area and therewith adhesion. Data obtained in the second experiment shows evidence for an exonuclease activity of Eap, degrading up to 2.5 kb linear DNA within 1 h. In contrast, circular plasmid DNA incubated with Eap was stable for the length of the

experiment. We suggest the DNA nuclease activity to be an adaptation to Neutrophil extracellular traps (NETs) which are primarily composed of DNA.

P.61 Pharmacological regulation of the Ca^{2+} homeostasis in the endoplasmic reticulum

Pick, T.[1], Dudek, J.[2], Jung, M.[2], Zimmermann, R.[2], Cavalié, A.[1]

[1]*Department of Experimental and Clinical Pharmacology & Toxicology, Saarland University, Homburg, Germany*

[2]*Department of Medical Biochemistry & Molecular Biology, Saarland University, Homburg, Germany*

Misfolded Proteins are usually degraded by proteasomes in the cytosol. Thus, such proteins have to be transported retrogradely out of the endoplasmic reticulum (ER), and this mechanism hampers ER overload and ER stress, which can lead to apoptosis. Accordingly, proteasome inhibitors lead to an accumulation of misfolded proteins in the ER. In order to analyze the functional effect of proteasome inhibitors on the Ca^{2+}-homeostasis, the cytosolic and ER-lumenal Ca^{2+}-concentration were measured in HEK-293-cells with Fura-2 and Förster resonance energy transfer (FRET), respectively. Our results indicate that proteasome-inhibitors cause an increase in the Ca^{2+}- leak of the ER and, hence, a dramatic change in the Ca^{2+}-homeostasis resulting in a fast Ca^{2+}-efflux. Likely, the regulation of Ca^{2+}-channels in the ER is potentially impaired by the treatment with proteasome inhibitors. Since the Sec61-complex has been shown to function as a Ca^{2+}-leak channel in the ER under certain conditions, we want to characterize the function of Sec61-complexes in a multiple myeloma cell model, the cell line RPMI 8226, which secretes large amounts of immunoglobulin G light chains. We hypothesize that the RPMI 8226 cells express high amounts of Sec61 proteins as it is expected to be needed for high protein secretion and, accordingly, these cells might be highly susceptible to proteasome inhibitors. The combination of the treatment with proteasome inhibitors and Ca^{2+}-measurements might pro- vide new insights on the function of the Sec61-complex under ER stress.

P.62 Fast and slow excitability in models of intracellular calcium release

S. Rüdiger

Institut für Physik, Humboldt-Universität zu Berlin, Berlin, Germany

Calcium dynamics are essential to a multitude of cellular processes. For many cell types, localized discharges of calcium through clusters of intracellular channels are building blocks to all spatially extended calcium signals. We discuss an emerging modeling approach based on the complex excitable dynamics of single clusters of IP3 receptor channels. We first present a model for calcium puffs using master equations for receptor states and differential equations for calcium in the cluster domain. We show that a transition to a slow modulation in the cluster dynamics results in a state consistent with many features of waves, including their long periods and refractoriness [1]. Our study suggests that waves are established by a random but time-modulated appearance of sustained release events, which have a high potential to trigger and synchronize release throughout the cell. We then present recent advances in modeling with continuous channel gating variables. We derive stochastic differential equations and show that the model reveals excitability similar to many classical cell systems [2]. In the deterministic limit, the model also predicts a bifurcation from mono- to bistability producing the slow dynamical component observed in calcium waves.

[1] M. Rückl, I. Parker, J.S. Marchant, Ch. Nagaiah, F.W. Johenning, S. Rüdiger, Modulation of elementary calcium release mediates a transition from puffs to waves, in preparation.

[2] S. Rüdiger, Excitability in a stochastic differential equation model for elementary calcium release, to appear in Phys. Rev E.

P.63 Cooperative motion of bacteria on soft surfaces

Benedikt Sabass

Complex Fluids Group, Princeton University, Princeton NJ, USA

Myxobacteria on surfaces propel themselves by two distinct macromolecular mechanisms, namely twitching and gliding. During collective migration, retraction of type-IV pili drives the twitching motion of the bacteria. In contrast, the "adventurous" motion of single bacteria is believed to be powered by intracellular motor complexes that produce thrust on transmembrane complexes, which is qualitatively similar to the gliding mechanism of Apicomplexans. Here, we use traction force microscopy to study bacterial forces during collective and adventurous migration. In particular, we ask how forces from individual bacteria are coordinated and how elastic properties guide the collective motion.

P.64 Shape transformation during endospore formation in bacteria

Anirban Sain

Physics Department, Indian Institute of Technology-Bombay, Mumbai, India.

In adverse conditions some bacteria like *Bacillus subtilis* form endospore, a spore which is engulfed by the mother cell itself and released into the environment when conditions are favorable. We construct a model to describe the dynamics of the membrane shape as sporulation proceeds. In wild type cells, following an asymmetric cell division a flat septum forms near one of the cell poles. Later the smaller cell rounds up and detaches itself from the cell wall of the mother cell to form a spore. We also examine the anomalous case when a bulge forms on the flat septum which eventually rounds up to form a vesicle.

The synthesis of peptidoglycan, material for the cell wall, also plays an important role during the process.

P.65 Living cell rheology: Fourier modes and spontaneous dynamic symmetry breaking

Mathias Sander and Albrecht Ott

Biological Experimental Physics, Saarland University, Saarbrücken, Germany

Active locomotion, tissue formation and wound healing involve mechanical activity of biological cells. This activity is based on the cytoskeleton, a dynamic biopolymer network that supports mechanical cell stress. Although the underlying biochemical elements have been investigated at lengths, to date we are still far from a predictive framework for cell mechanical behaviour. Depending on stimuli, living cells show a variety of rheological changes including sti#ening and softening. Here we use the Cell Monolayer Rheology-technique (CMR) [1] in combination with Fourier Transform Rheology (FTR) to investigate nonlinearities in the cell mechanical response of 3T3 fibroblast cells quantitatively. We observe spontaneous, dynamic symmetry breaking in response to symmetric driving. We suggest that this is due to an active cytoskeletal response. We show that simple constitutive equations based on nonlinear elasticity and plasticity describe the measurements surprisingly well, except for the asymmetry.

[1] Fernandez P, Ott A, Aksel N, Heymann L, Pullarkat P A: Shear rheology of a cell monolayer. New Journal of Physics 9, 419 (2007).

P.66 Competition for binding leads to ultrahigh oligonucleotide molecular recognition specificity

Marc Schenkelberger[1,2], Christian Trapp[1], Timo Mai[1,4] and Albrecht Ott[1,3,4]

[1]*Biological Experimental Physics, Saarland University, Saarbrücken, Germany*

[2]*Physics Department, University of Minnesota, Minneapolis MN, USA*

[3]*Institut Curie, Paris, France*

Living matter requires molecules to find their matching binding partners within a mixture of many competitors. For non-interacting molecules in thermal equilibrium, the specificity of this recognition process is given by the Boltzmann factor of their difference in binding free energy, and this is believed to also apply to oligonucleotide hybridization (binding). Here we show that competition can increase DNA oligonucleotide molecular recognition specificity by a few orders of magnitude. In our experiments the competing molecules differ by only a single mismatch, and a change in its position may change or abolish this increase in specificity. We attribute our observation to an entropic energy barrier that results if both competitors bind to the same probe in a triplex configuration. This barrier affects the weaker binding competitor much more strongly, and the triplex configuration is hardly populated. Numerical assessment of the corresponding free energy landscape, expressed as a Landau mean field development, quantitatively yields the experimental result. Our results demonstrate how, in spite of the Boltzmann law, high molecular specificity can be reached in thermal equilibrium at the simple thermodynamic cost of introducing the competitor.

P.67 Spontaneous autocatalysis in a primordial broth

Sabrina Scherer, Eva Wollrab, Albrecht Ott

Biological Experimental Physics Department, Saarland University, Saarbrücken, Gemany

Driven non-linearities lead to pattern formation. Here we study the dynamics of a complex chemical system, driven by electric discharge that forms from a gas mixture of methane and ammonia in the presence of water. Using real-time mass spectrometry, we observe the generation of a primordial broth composed of thousands of different molecules in a mass range from 50 to 1000 Dalton. The temporal development of the primordial broth reveals the spontaneous emergence and disappearance of oligomers. Strong non-linearities are required for these aperiodic chemical oscillations. The phenomenon is robust against different gas compositions and concentrations, temperatures and many details of the experimental set-up. We analyze the chemical composition of the solution by NMR and different types of (high-resolution) mass spectrometry to find amine-rich carbon chains, polyethylene glycol (PEG) based surfactants and other catalysts. We find that oxidation and doping with small amounts of an active broth can trigger the production of the oligomers. We suggest that surface active molecules lead to phase transfer catalysis in the oil/water mixture and self-organize to a spontaneously emerging autocatalytic network.

P.68 The passive Ca^{2+}-efflux from the endoplasmic reticulum through the Sec61 complex: Regulation and physiological consequences

Stefan Schorr[1], Nico Schäuble[1], Martin Jung[1], Adolfo Cavalié[2] and Richard Zimmermann[1]

[1]*Medical Biochemistry and Molecular Biology, Saarland University, Homburg, Germany,*

[2]*Pharmacology and Toxicology, Saarland University, Homburg, Germany*

One third of all proteins encoded in a eukaryotic genome have to be translocated across or inserted into the membrane of the endoplasmic reticulum (ER) in a process mediated by the Sec61-complex. The heterotrimeric Sec61-complex forms a hydrophilic channel in the ER membrane which transiently allows the passage of calcium ions (Ca^{2+}) *in vivo*, especially after termination of protein translocation. In mammalian cells, the ER serves as the major storage organelle for Ca^{2+}. Thus the cell has to manage the opposing tasks of the frequent opening of the translocation pore and simultaneously preserving the ion gradient between cytosol (50 nM Ca^{2+}) and ER-lumen (800 µM Ca^{2+}). We found that the ER luminal Hsp70-type chaperone BiP plays a central role in Sec61 channel gating. To study the role of BiP and its cofactors in Ca^{2+}-efflux control we employ siRNA mediated gene silencing in combination with live cell calcium imaging.

We identified a conserved binding site for BiP in the lumenal loop 7 of Sec61α which is critical in the channel regulation. The regulation involves a classical substrate recognition/binding circle of BiP and, therefore, depends on the action of an Hsp40 cochaperone and nucleotide exchange factors. We identified the two soluble Hsp40 proteins ERj3 and ERj6 as the critical components controlling the function of BiP during the regulation of the Ca^{2+}-leakage through Sec61α. Interestingly, the three ER-membrane bound Hsp40 proteins (ERj1,Sec63,ERj7), which control BiP functions during protein translocation, are not involved in the Ca^{2+}-leakage control, indicating functional specialization in the Hsp40 network. The notion of an involvement of a functional ATPase cycle of BiP is further strengthened by the observation that the simultaneous depletion of Sil1 and Grp170,

the two redundant nucleotide exchange factors of BiP, phenocopy the effect of BiP depletion.
We propose a model in which ERj3 and ERj6 recruit BiP to the loop 7 and, subsequently, stimulate its ATPase activity to ensure proper binding to Sec61 in order to limit the Ca^{2+}- efflux. If this mechanism is compromised, the increased Ca^{2+}-efflux leads to diverse cellular stress responses and triggers apoptosis especially in highly secretory active cells. This view is supported by the fact that mice carrying a mutation in the BiP binding motif in Sec61 as well as ERj6-/- mice develop diabetes type II due to apoptosis of β-cells in the pancreas.

P.69 Less-invasive transverse aortic banding in mice for hypertrohy studies

Laura Schröder, Qinghai Tian, Elisabeth Kaiser, Peter Lipp, Lars Kaestner

Institute for Molecular Cell Biology, Saarland University, Homburg, Germany

Cardiac hypertrophy is a critical accompanying symptom associated with many cardiovascular diseases. To study the multifactorial processes involved, the use of surgically induced in vivo models is often unavoidable, especially of mice undergoing aortic banding. In the conventionally used transverse aortic constriction (TAC) model, the survival rate is a serious problem, especially regarding rare or expensive genetically modified mice. For our hypertrophy models, we therefore use the minimally invasive aortic banding, MTAB (Martin et al. 2012), with modifications. In this surgery, the access is located cranially of the sternum, avoiding the entering of the pleural space. This both circumvents the risk of a lethal pneumothorax and pleural infection, as well as artificial ventilation and intubation, and reduces the time under anesthesia. Thus, it is less harmful to the animal. After surgery, the hypertrophic process can be in vivo- imaged by cardiac ultrasound, to assess the development of cardiac morphology and performance during hypertrophy. Finally, cardiac myocytes are isolated, and used in further investigations, as imaging the T-tubules. They are stained with di-8-ANEPPS, a membrane-selective fluorescent

dye, to measure the regularity of the transverse tubules (T-Tubules) by using confocal microscopy techniques. After 7d of TAC in situ, there is a significant difference in T---Tubular regularity in isolated cardiac myocytes in mice.

P.70 Intermittent search and transport strategies in spherical domains

Karsten Schwarz and Heiko Rieger

Theoretical Physics, Saarland University, Saarbrücken, Germany

The successful application of effcient search strategies is one of the most important needs in biology and human behavior. It is observed on all length scales of life and in all kinds of complexity. A frequently used way of modelling real search is a so-called intermittent search strategy [1]. The searcher switches between phases of fast directed ballistic motion, in which it cannot recognize a target, and phases of slow diffusion for detecting a target. A biological relevant example, where this way of describing the real motion fits very well, is intracellular transport. With the help of an event driven kinetic Monte Carlo method [2] we study the efficiency of this search strategies for different scenarios like narrow escape problems and predator-prey situations. The goal is always to find the strategy which minimizes the Mean First Passage Time (MFPT) as a function of the freely choosable parameters.

[1] C. Loverdo, O. Benichou, M. Moreau, R. Voituriez, Enhanced reaction kinetics in biological cells, Nature Physics 4, 134 (2008).

[2] K.Schwarz and H. Rieger, Efficient kinetic Monte Carlo method for reaction-diffusion problems with spatially varying annihilation rates, J. Comp. Phys. 237, 396-410 (2013).

P.71 Mechanistic Role of Q_{bc}-SNAREs in vesicular cargo release

Ahmed Shaaban[1], Ali Harb[1], Dieter Bruns[2], and Ralf Mohrmann[1]

[1]ZHMB Nachwuchsgruppe Physiologie, [2]Institut für Physiologie, Saarland University, Homburg, Germany

The regulated fusion of transport vesicles with the plasma membrane underlies cellphysiological processes like secretion of signaling molecules and surface delivery of receptors that are essential for intercellular communication. During exocytosis the formation of ternary SNARE complexes out of Syntaxin-1A, Synaptobrevin-2, and SNAP-25 constitutes a central mechanistic step that is believed to result in membrane merger and cargo release. In contrast to its membrane-anchored binding partners that possess only a single SNARE motif, SNAP-25 (Q_{bc}-SNARE) contains two α-helical SNARE domains (Q_b and Q_c) and is attached to the plasma membrane via palmitoylation. Interestingly, the functional implications of this conspicuous structural arrangement in SNAP-25 have not been fully understood yet. Especially, the question whether the two SNARE motifs must be physically linked to allow for normal exocytosis has been controversially discussed. To investigate the exact mechanistic role of the connecting linker in SNAP-25 we co-expressed separate Q_b and Q_c motifs with adjoining sequences required for palmitoylation and proper localization at the plasma membrane. While electrophysiological recordings in neuroendocrine cells demonstrated a full reconstitution of exocytosis by expression of full-length SNAP25 in SNAP25$^{-/-}$ ells, the co-expression of the separate fragments completely failed to support hormone secretion under the same experimental conditions, stressing the importance of a connection between both domains. Indeed, expression of either SNAP-25 SNARE motif alone inhibited secretion in wildtype cells, with the C-terminal fragment having the most severe effect. However, complete removal of the linker abolished inhibition by the C-terminal fragment, which suggests that the linker itself may act as a nucleation site for association with other SNARE partners. Further experiments are planned to extend our structure-function analysis.

In complementing experiments we have also investigated the role of Q_{bc}-SNAREs in membrane insertion of neurotransmitter receptor in hippocampal neurons. For this purpose we have analyzed the trafficking of fluorophor (ecliptic GFP)-tagged glutamate-receptors using *total internal reflection fluorescence* microscopy. Our preliminary results have demonstrated that SNAP-25 is also an essential component of the molecular machinery employed to deliver receptors onto the cell surface.
Taken together, our results indicate that SNAP-25 is a structurally specialized SNARE protein that is involved in different forms of exocytotic cargo release.

P.72 Anomalous diffusion of self-propelled particles on directed random networks

M. Reza Shaebani, Zeinab Sadjadi, Heiko Rieger, and Ludger Santen

Theoretical Physics, Saarland University, Saarbrücken, Germany

We study the influence of structural characteristics of filamentous networks, or equivalently the stepping strategy in continuous space, on the transport properties of a random walker. A general master equation formalism is developed to investigate the persistent motion of self-propelled particles, which enables us to identify the key parameters and disentangle their contributions to the transport process. An anomalous diffusive behavior emerges, depending on the choice of the persistency of the walker and the anisotropy and heterogeneity of the structure or stepping properties. We establish the existence of up to three different regimes of motion, and determine the phase diagrams of the behavior. We verify that the crossover times between different regimes as well as the long-term diffusion coefficient can be enhanced by a few orders of magnitude within the biologically relevant range of control parameters. The analytical predictions are in excellent agreement with simulation results.

P.73 Molecular mechanisms of exocytosis of large dense core vesicles in dorsal root ganglion neurons

Ali Shaib, Margarete Klose, Jens Rettig, Barbara Niemeyer and Ute Becherer

Physiology Institute, Faculty of Medicine, Saarland University, Homburg, Germany

Dorsal root ganglion (DRG) neurons transmit sensory information to the central nervous system through glutamatergic synaptic transmission that can be modulated by peptide secretion. They are contained in large dense core vesicles (LDCVs) and can be as diverse as Calcitonin gene-related peptide, neuropeptide Y, substance P, etc. LDCVs are exocytosed in response to specific stimulus in afferent terminals along with synaptic vesicles (SVs) and at the cell somata. The release machinery of these LDCVs is poorly understood. Thus, we investigate the role of CAPS1 and CAPS2 which are known priming factors in chromaffin cells. Using PCR, we verified that both CAPS isoforms are expressed. With total internal fluorescence reflection microscopy, we visualize in real time the release of LDCVs that are labeled through Lenti virus driven expression of NPY-Venus. We found that the secretion in WT DRG-neurons co over-expressing CAPS2b was 3 fold larger than in control, indicating that CAPS2b can be a priming factor in DRG-neurons. We are currently verifying this finding by measuring the LDCV exocytosis in CAPS1 and 2 double KO DRG neurons. We also examine the role of CAPS2b in SV priming. For this we have established a DRG-dorsal horn neurons co-culture to allow synapse formation. Additionally, we designed a Lenti virus encoding for vGlut tagged to the pH sensitive fluorescent protein mNectarin to specifically label SVs and visualize their exocytosis. With these tools we are investigating whether CAPS2b plays a differential role in priming of LDCV vs. SV.

P.74 Probing the role of spatial anisotropy and heterogeneity in contractility and adhesion distribution during cell steering

Soumya S S[1], Subodh Kolwankar[1], Edna George[2], Santanu K. Basu[3], Shamik Sen[2], and Mandar M. Inamdar[1]

[1]*Department of Civil Engineering, Indian Institute of Technology Bombay, Mumbai, India*

[2]*WRCBB, Department of Biosciences and Bioengineering, Indian Institute of Technology Bombay, Mumbai India*

[3]*Department of Chemical Engineering, Indian Institute of Technology Bombay, Mumbai, India*

Cell motility is central to various processes including embryogenesis and wound healing. For efficient migration, close temporal and spatial co-ordination between the cytoskeleton and the focal adhesions is essential, with cell shape stability maintained at shorter timescales. Since both the cytoskeleton and the adhesions contribute to motility, their relative contributions to a cell's frequent directional changes remain unclear. Tensional homeostasis, or force balance between the cytoskeleton and the focal adhesions, can be gauged using the trypsin de-adhesion assay wherein, cell retraction is tracked upon rapidly severing cell-matrix contacts using the enzyme trypsin. Upon incubation with trypsin, in addition to the sigmoidal retraction kinetics observed in a variety of adherent cells, 3T3 fibroblasts were found to exhibit a combination of translation and rotation while rounding up. Such motions may arise from a combination of anisotropy in cell contractility (or, prestress) and cell-matrix adhesions, and may be associated with frequent directional changes associated with random cell motility. In this study, we propose a simple theoretical model, which can replicate the above-mentioned cell dynamics, and evaluate the relative contributions of anisotropy in contractility and bond distribution. The cell represented by a circle of unit radius is connected to the substrate by continuous bonds, with both cell and substrate modeled as continuous, homogeneous, isotropic visco-elastic Kelvin-Voigt materials. Various combinations of bond distribution and

prestress were applied to the cell and then using finite element analysis, its translation and rotation upon trypsin assisted de-adhesion were examined in each case. The results of our study indicate that cellular translation arises due to partial asymmetry about any axis in the bond distribution and (or) bond strength, with contractility dictating the magnitude of the final movement. We also show that while asymmetry in bond distribution causes only cell translation, a combination of asymmetric bond distribution and non-uniform contractility are required for translation and rotation, and may guide cell migration.

[1] S. Sen and S. Kumar, Cell. and Mol. Bioeng. 2, 218 (2009).

P.75 Influence of surface and subsurface properties on the structure and activity of adsorbed bactericidal proteins

Christian Spengler[1], Christian Kreis[1], Stéphane Mesnage[2], Hendrik Hähl[1], Simon Foster[2], Karin Jacobs[1]

[1]*Experimental Physics, Saarland University, Saarbrücken, Germany*

[2]*University of Sheffield, The Krebs Institute, Department of Molecular Biology and Biotechnology, Sheffield, United Kingdom*

Protein adsorption is the first step in biofilm formation: Protein films serve as a conditioning layer that enables and affects the attachment of bacteria and other organisms. Hence, the understanding and control of protein layers is an important task that is relevant to life sciences and engineering. Previous studies revealed that the structure and density of adsorbed proteins and the adhesion force of bacteria depend on both the surface properties and the subsurface composition of the adsorbent material [1,2]. These findings raise the question whether or not the activity of adsorbed proteins is also influenced by the properties of the underlying material. In this study, we investigate how the activity - the bactericidal effect - of adsorbed lysozyme and lysostaphin is affected by surface properties. The activity is thereby characterized by measuring

the turbidity of a very sensitive protein assay containing purified peptidoglycan.

[1] Hähl et al., Langmuir 28, 7747-7756 (2012).

[2] Loskill et al., Langmuir 28, 7242-7248 (2012).

P.76 The role of vimentin in cell migration under confinement

Stankevicins L., Terriac E., Lautenschläger F.

Experimental Physics, Saarland University, Saarbrücken, Germany

Cell migration is a fundamental biological process which requires an active cytoskeleton remodelling. During migration, cells often perform long distance displacements passing through obstructive 3D tissues, constrictions, or even in between other cells and therefore continuously need to adjust their shape. The intermediate filaments are the most stretchable components of the cytoskeleton and are organized as a network comprising both cytoplasmatic and nuclear proteins. It is not well established if and how alterations in the extracellular matrix can have an impact in intermediate filaments organization and cell movement. Considering this scenario, we subjected cells to confinement using microfabricated channels with the objective to analyse cell migration and the cytoplasmatic intermediate filament vimentin network. In this given context we analysed two models of cell migration- the low adherent amoeboid performed by Jurkat cell lines and the mesenchymal migration using RPE-1 cells. Both cell lines were stable transfected with wild type vimentin coupled with GFP. Cell movement in the channels was recorded by fluorescence and phase contrast video microscopy for 24 hours under controlled humidity and temperature. The acquired data was analysed by a custom-built cell tracking software. The analysed parameters during both amoeboid and mesenchymal cell migration were the speed, persistence as well as the localization of vimentin network and cell morphology (length, shape, symmetry, nuclear position, nuclear shape).

P.77 Molecular and functional characterization of Munc119 in photoreceptor synapses

Shweta Suiwal, Karin Schwarz, Frank Schmitz.

Department of Anatomy, Cell Biology and Developmental Biology, Saarland University, Homburg, Germany

Ribbon synapses are high performance synapses built by certain non-spiking sensory neurons, e.g. photoreceptors, retinal bipolar cells and inner ear hair cells. They are characterized by high rates of synaptic vesicle exocytosis, coordinated multivesicular release, exquisite sensitivity over a wide dynamic range of signaling, including both phasic and tonic components. The main site of exocytosis is characterized by unique presynaptic structures, the synaptic ribbons, which are associated with large numbers of synaptic vesicles. Ribbon associated vesicles are primed for exocytosis and provide the active zone with a large pool of release-ready vesicles [2, 3]. Munc119 interacts with the synaptic ribbon protein RIBEYE [1]. Munc119 (also designated as RG4) is a mammalian ortholog of the *Caenorhabditis elegans* protein unc119. Munc119/RG4 is related to the prenyl-binding protein PrBP/δ and expressed at high levels in photoreceptor ribbon synapses. It is essential for vision and synaptic transmission in photoreceptor ribbon synapses by unknown molecular mechanisms. In order to analyze the function of a protein, identification of molecular interaction partners is important. Purpose of present project is to identify and characterize new interaction partners of Munc119 which could provide more insight on the functional significance of the ribbon synapse.

[1] Alpadi, K., Magupalli, V.G., Käppel, S., Köblitz, L., Schwarz, K., Seigel, G.M., Sung, C.H., Schmitz, F (2008). Journal of Biological Chemistry **283**, 26461-7.

[2] Magupalli, V.G., Schwarz, K., Alpadi, K., Natarajan, S., Seigel, G.M and Schmitz, F (2008) Multiple Ribeye-Ribeye interactions create a dynamic scaffold for the formation of synaptic ribbon. J. Neurosci. **28**, 7954-7967.

[3] Schmitz, F., Königstorfer, A and Südhof, TC (2000) RIBEYE, a component of synaptic ribbons: a proteins journey through evolution provides insight into synaptic ribbon function. Neuron **28**, 857-872.

P.78 A microfabricated modular strategy for probing the impact of physical cues on adhesive cells

Chiara Tamiello, Carlijn C.V. Bouten, Frank P.T. Baaijens

Department of Biomedical Engineering, Eindhoven University of Technology, Eindhoven, The Netherlands

Adhesive cells sense and continuously respond to the physical cues of the surrounding (micro) environment. This interaction, crucial for cell structural and functional integrity, involves the central activity of the actin cytoskeleton, which is continuously reorganized to achieve this function [1]. However, understanding cell behavior in response physical properties of the environment is far from complete.
Here we devise a modular strategy for investigating the synergistic and collective effect of stiffness, stiffness anisotropy and strain on cell behavior. The modules involve arrays of microfabricated elastomeric microposts characterized by different heights (stiffness), cross-sections (stiffness anisotropy) and incorporated into a stretching device (strain).
Using this strategy, we have investigated actin cytoskeleton response of vascular–derived cells (HVSC) seeded on microposts of two heights, with and without stiffness anisotropy and subjected to pure uniaxial cyclic stretch (10%, 0.5Hz for 19 hours). Our data show that in response to this strain regime, strain avoidance response governs actin cytoskeleton behavior in case of stiffness isotropy. On the other hand, when stiffness anisotropy is presented to HVSC, the strain avoidance response of the actin cytoskeleton depends on the stiffness of the microposts.
Taken together, our strategy represents a powerful tool for obtaining mechanistic insights in the impact of physical cues onto cell response.

[1] Fletcher D. A. & Mullins R. D., Nature 463, 485–492 (2010).

P.79 Aster migration in Sea Urchin eggs studied by 3D tracking and cell shape manipulation

Hirokazu Tanimoto[1], Akatsuki Kimura[2], Nicolas Minc[1]

1Institut Jacques Monod, Paris, France

2National Institute of Genetics, Mishima, Japan

Immediately after fertilization, radial microtubules (MTs) start growing from the centrosome brought inside the egg by the sperm. This sperm MT aster continuously grows in size and concomitantly propels itself to the center of the large egg. This centration process is thought to be driven by the integration of individual MT forces which scale with MT length [1-3]. To date however, very little is known on the basic self-organization principle regulating dynamics and geometry sensing.

We here study aster centration process using three dimensional confocal microscopy and image analysis. We find that the growing aster migrates with a constant velocity in most of the centration period, with impressive persistency towards the cell center. We investigate effects of cell geometry by manipulating eggs in micro-fabricated chambers, and cell size by cutting eggs with micropipettes, on aster trajectories and speeds. We find that the velocity is independent on boundary geometry and absolute time of migration. Numerical simulations can predict the dependence of aster trajectories on cell geometrical boundaries, and suggest that aster velocity may primarily be determined by MT polymerizing rate, rather than absolute force.

[1] Wuhr et al., Curr. Biol. (2011).

[2] Kimura and Kimura, PNAS (2011).

[3] Minc et al., Cell (2011).

P.80 Modeling the instability of an expanding cellular mono-layer during collective cell motion

Victoria Tarle

Weizmann Institute of Science, Rehovot, Israel

Collective motion of cells has been of interest for researchers for many years, as it is involved in a variety of key processes, such as wound healing, tumor invasion, and embryogenesis. In-vitro experiments conducted to study the movement of cell mono-layers upon confinement removal, revealed the spontaneous formation of finger-like columns of cells, with “leader cells” at their tips. The leader cells have distinguishable motility properties. Here we have implemented a curvature-motility feedback for the cells at the edge of the layer, and combined this with a consistent description of cell motility that was previously used to describe the properties of the bulk.
Our particle-based simulation, modeling the cells as single points, shows the spontaneous formation of the fingers, which we then compare to those observed in experiments. The model is based on several theoretical approaches: An interaction potential (attractive and repulsive) between neighboring cells, and treating the cells as self-propelled particles, which have a certain degree of velocity correlation (Vicsek Model). The model allows us to explore the effects of these different components on the formation and properties of the fingers.

P.81 Hydrophobic interaction governs unspecific adhesion of staphylococci: a single cell force spectroscopy study

Nicolas Thewes[1], Peter Loskill[1], Philipp Jung[1], Henrik Peisker[2], Markus Bischoff[2], Mathias Herrmann[2] and Karin Jacobs[1]

[1]*Experimental Physics, Saarland University, Saarbrücken, Germany*

[2]*Institute of Medical Microbiology and Hygiene, Saarland University, Homburg, Germany*

Unspecific adhesion of bacteria is usually the first step of biofilm formation on abiotic surfaces, yet it is unclear up to now which forces are governing this process. Alongside long-ranged van der Waals and electrostatic forces, short-ranged hydrophobic interaction plays an important role. To characterize the forces involved during approach and retraction of an individual bacterium to and from a surface, single cell force spectroscopy is applied: A single cell of the apathogenic species S. carnosus isolate TM300 is used as bacterial probe. With the exact same bacterium, hydrophobic and hydrophilic surfaces can be probed and compared. We find that as far as 50 nm from the surface, attractive forces can already be recorded, an indication of the involvement of long-ranged forces. Yet, comparing the surfaces of different surface energy, our results corroborate the model that large, bacterial cell wall proteins are responsible for adhesion, and that their interplay with the short-ranged hydrophobic interaction of the involved surfaces is mainly responsible for adhesion. The ostensibly long range of the attraction is a result of the large size of the cell wall proteins, searching for contact via hydrophobic interaction. The model also explains the strong (weak) adhesion of S. carnosus to hydrophobic (hydrophilic) surfaces.

P.82 Ca^{2+} leak channels in the endoplasmic reticulum (ER)

Tinschert, R.[1], Vieró, C.[1], Dudek, J.[2], Jung, M.[2], Zimmermann, R.[2], Cavalié, A.[1]

[1] Department of Experimental and Clinical Pharmacology & Toxicology and [2]Department of Medical Biochemistry & Molecular Biology, University of Saarland, Homburg, Germany

Ca^{2+} functions as a universal and versatile ion-messenger in a variety of cellular processes, thereby acting as a second-messenger. The intracellular Ca^{2+} level is tightly regulated by the combined action of organelles, pumps (e.g. SERCA), exchangers (e.g. NCX), and diverse Ca^{2+}- selective ion-channels. In the resting state, $[Ca^{2+}]_i$ is approx. 50-100 nM and rises up to ca. 1 µM under the action of Ca^{2+}-mobilizing agents. These intracellular Ca^{2+}-transients and global $[Ca^{2+}]_i$ signals are decoded in terms of speed, amplitude, and duration by Ca^{2+}-binding proteins like calmodulin. Thereby, the Ca^{2+}-efflux from intracellular stores, i.e. the sarcoplasmic reticulum (SR) in contractile cells, or the endoplasmic reticulum (ER) in other cell types, respectively, plays an important role in cellular processes such as the electromechanical-coupling in muscle cells. $[Ca^{2+}]$ER/SR is approx. 100-800 µM and the Ca^{2+}-efflux from ER/SR is mainly mediated by Ca^{2+}-channels such as IP_3-, and ryanodine-receptors. The SEC61-complex, a protein conducting channel in the ER, which is responsible for the translocation of proteins into the lumen of the ER, was also shown to function as a "Ca^{2+}-leak channel" under certain conditions. Aim of the present study is to investigate the mechanism and function of the human SEC61-complex as a "Ca^{2+}-leak channel" in different model systems (dog-pancreas, and cell culture lines) by using electrophysiological techniques. Here we use a novel automated, chip-based planar patch-clamp- system called "port-a-patch".

P.83 Furrow constriction in animal cell cytokinesis

Hervé Turlier[1,2], Basile Audoly[3], Jacques Prost[1,4], Jean-François Joanny[1]

[1] *Physicochimie, Institut Curie, Centre de Recherche, Paris, France*

[2] *European Molecular Biology Laboratory, Heidelberg, Germany*

[3] *Institut Jean Le Rond d'Alembert, Université Pierre et Marie Curie, Paris, France*

[4] *ESPCI-ParisTech, Paris, France*

Cytokinesis is the process of physical cleavage at the end of cell division; it proceeds by ingression of an actomyosin furrow at the equator of the cell. Its failure leads to multinucleated cells and is a possible cause of tumorigenesis. Here, we calculate the full dynamics of furrow ingression and predict cytokinesis completion above a well-defined threshold of equatorial contractility. The cortical actomyosin is identified as the main source of mechanical dissipation and active forces. Thereupon, we propose a viscous active non-linear membrane theory of the cortex that explicitly includes actin turnover and where the active RhoA signal leads to an equatorial band of myosin overactivity. The resulting cortex deformation is calculated numerically and reproduces well the features of cytokinesis such as cell shape and cortical flows toward the equator. Our theory gives a physical explanation of the independence of cytokinesis duration on cell size in embryos. It also predicts a critical role of turnover on the rate and success of furrow constriction. Scaling arguments allow for a simple interpretation of the numerical results and unveil the key mechanism that generates the threshold for cytokinesis completion: cytoplasmic incompressibility results in a competition between the furrow line tension and the cell poles surface tension.

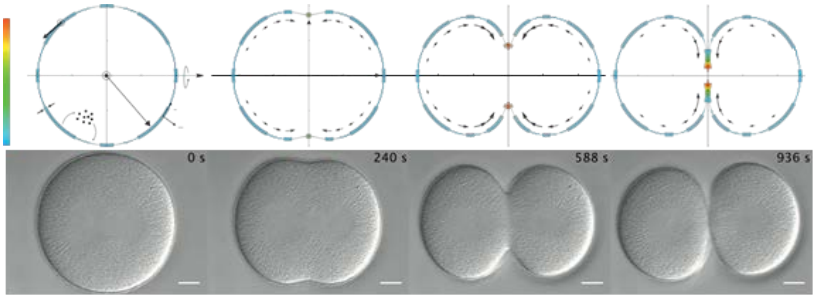


Fig. 1: Comparison of numerical & experimental successive snapshots of furrow constriction

[1] H. Turlier, B. Audoly, J. Prost and J-F. Joanny, Biophysical Journal 106(1) 2014.

P.84 Heterologous expression and characterization of N-terminal domains of NOX5: a calcium dependent NADPH oxidase

Rama Ramesh Vandanapu, C. Roy. D. Lancaster

Department of Structural Biology, Institute of Biophysics and Centre of Human and Molecular Biology (ZHMB), School of Medicine, Saarland University, Homburg, Germany

NADPH oxidases (NOX) are enzymes involved in transmembrane electron transfer to molecular oxygen, thereby generating superoxide radicals. Unlike other members of the family, NOX5 binds Ca^{2+} via its N-terminal domain and does not require cytoplasmic subunits for activation. Calcium binding has been shown to induce intramolecular interactions between the N-terminal and the C-terminal domains in the cytosol, triggering transmembrane electron flow and hence enhancing production of superoxide. We produced the recombinant N- terminal domains of NOX5 isoforms and characterized the conformational changes they undergo upon calcium binding. Our results show that different NOX5 isoform N-terminal domains undergo distinct

conformational changes in the presence of calcium. To gain deeper understanding of these conformational changes, we modelled solution structures of these isoforms based on small-angle X-ray scattering (SAXS) studies in the presence and absence of calcium. These different isoforms also have different affinities for calcium. The results will be discussed.

P.85 Activity Driven Fluctuations in Living Cells

É Fodor[1], W. W. Ahmed[2], T. Betz[2], M. Bussonnier[2], N. S. Gov[3], M. Guo[4], V. Mehandia[5], D. Riveline[5], P. Visco[1], D. A. Weitz[4], F. van Wijland[1]

[1]*Laboratoire MSC, Université Paris Diderot and CNRS, France*
[2]*Laboratoire P.C.C., Institut Curie, Paris, France*
[3]*Department of Chemical Physics, Weizmann Institute of Science, Israel*
[4]*School of Engineering and Applied Sciences, Harvard University, Cambridge, MA.*
[5]*Laboratory of Cell Physics, Universite de Strasbourg and CNRS, France*

Present day imaging techniques allows for particle tracking with unprecendented statistics. It can further be coupled to active microrheology experiments by means of optical trapping devices. Accessing the characteristics of the motion—whether free or driven—is exploited in a variety of experimental living media, ranging from isolated myoblasts or glial cells, to oocytes and epithelial tissues, leading to an explosion of data. While these experimental data are now of very high statistical quality and abundant, the original motivation, namely quantitatively understanding the role of active processes in shaping fluctuations in these living media, remains an elusive goal. In order to address this frustating contrast between experimental progress and physical understanding, we propose a model for the dynamics of a micron-sized probe embedded in an active medium, where both thermal fluctuations and nonequilibrium activity compete. The model comprises confining harmonic potential, describing the elastic cytoskeletal matrix, which undergoes random active hops, which represent the active forces and rearrangements within the cell. We describe the statistics of the

probe motion and we bring forth quantities affected by the nonequilibrium activity. By fitting our predictions with experimental results we are able to provide numerical values for the parameters characterizing motors activity, such as the typical time-scale of the activity and the amplitude of the active fluctuations. Finally, we successfully apply and extend our model to describe nonequilibrium fluctuations of different experimental setups, such as beads injected in living cells, vesicles inside oocytes and tricellular junctions in a cell monolayer.

P.86 Role of actin filaments in correlating nuclear shape and cell spreading

Renu Vishavkarma[1], Swetavalli Raghavan[1], Abhijit Majumder[2], Jyotsna Dhawan[2], and Pramod A. Pullarkat[1]

[1]*Raman Research Institute, Bangalore, India*

[2]*InStem, National Centre for Biological Sciences, Bangalore, India*

It is well known that substrate properties like stiffness and adhesivity influence cell morphology, cell dynamics, proliferation and even cell fate. Recent experiments show that cell morphology influences nuclear geometry and hence gene expression profile in case of pluripotent cells. The mechanism by which surface properties regulate cell and nuclear properties is only beginning to be understood. Direct transmission of forces from the adhesion sites to the cellular nucleus and biochemical signaling are involved in this process. Here, we investigate the formal aspect by studying the correlation between cell spreading and nuclear deformation using Mesenchymal stem cells under a wide variety of conditions. It is observed that a robust quantitative relation holds between the cell and nuclear projected areas, irrespective of how the cell area is modified or when various cytoskeletal or nuclear components are perturbed. By studying the role of actin stress fibers in compressing the nucleus we propose that nuclear compression by stress fibers can lead to enhanced cell spreading due to interplay between elastic and adhesion factors. The significance of myosin- II in

regulating this process is also explored. By applying a normal compression using a lens and a semi quantitative model, we establish that the nuclear elasticity controls the extent of cell spreading on a substrate and in order to increase the spreading, cell modifies the traction and stress fiber structures to compress the nucleus.

[1] Yeung T, Georges PC, Flanagan LA, Marg B, Ortiz M, et al. Effects of substrate rigidity on cell morphology, cytoskeletal structure and adhesion. Cell motility and the cytoskeleton 60: 24—34 (2005).

[2] Engler AJ, Sen S, Sweeney HL, Disher D Matrix elasticity directs stem cell lineage specification. Cell 126: 677—689 (2006).

[3] McBeath R, Pirone DM, Nelson CM, et al. Cell shape, cytoskeletal tension and RhoA regulates stem cell lineage commitment. Developmental cell 6: 483-495 (2004).

[4] Khatau SB, Hale CM, stewart-Hutchinson P, Patel MS, et al. A perinuclear actin cap regulates nuclear shape. PNAS 106: 19017-19022 (2009).

[5] Versaeval M, Grevesse T, Gabriele S. Spatial coordination between cell and nuclear shape within micropatterened endothelial cells. Nat Commun 3: 671 (2012).

[6] Tatara Y On compression of rubber elastic sphere over a large range of displacement—Part 1: theoretical study. J Eng Matter Technol 113: 285-291 (1991).

[7] Tatara Y, Shima S, Lucero JC. On compression of rubber elastic sphere over a large range of displacement—part 2: Comparison of theory and experiment. J Eng Matter Techol 113: 292-295 (1991).

P.87 Motor-driven biological transport under confinement

Tobias Weber, M. Reza Shaebani, and Ludger Santen

Theoretical Physics, Saarland University, Saarbrücken, Germany

The nervous system of highly developed animals consists of a network of cells which pervade the whole organism. Being connected by axons and dendrites, single nerve cells are able to transmit and receive information via electric signals. Apart from this, axons and dendrites

serve as channels for many transport processes taking place on microtubules in their interior. These transport processes are crucial for the function and survival of the neuron. In the present work, transport processes are simulated in a cylindrical shape, representing the axon, which contains a dynamic microtubule network. We consider the transport of cargoes which are driven by teams of molecular motors. Asymmetric motor configurations are considered such that a given motor-cargo complex performs a biased motion along the MT bundle despite its ability to move both directions. Numerical simulations show that for systems with oppositely biased cargoes a dynamic phase transition from a homogeneous to a jammed phase exists which is controlled by the asymmetry of the cargo motion. We also apply the theoretical model to realistic axonal geometries which we reconstruct from microscopic images.

P.88 Vascularization patterns and fluid flow in growing tumors

M. Welter and H. Rieger

Theoretical Physics, Saarland University, Saarbrücken, Germany

Growing tumors remodel the vascular network by generating new blood vessels (angiogenesis), by co-opting already existing vessels from the arterio-venous vasculature of the surrounding healthy tissue and by vessel regression. We want to understand the physical determinants of the emerging tumor vascularization patterns and the characteristics of the resulting blood and interstitial fluid flow. For this purpose we develop a theoretical model [1] combining a dynamically evolving and blood flow carrying pipe network with a non-linear growth process, intercommunicating via oxygen, nutrient and growth factor fields. With the help of this model we discuss mechanisms leading to tumor compartmentalization, hot spot formation, oxygen distribution patterns, vascular blood flow characteristics, and interstitial fluid flow features including their consequence for a successful drug delivery within the tumor.

[1] M. Welter, H. Rieger: PLoS ONE 8, e70395 (2013).

P.89 Intracellular Ca^{2+} content and the regulation of phosphatidylserine exposure in human red blood cells

Mauro C. Wesseling, Henri Huppert, Benjamin Hanf, Lisa Wagner-Britz, Ingolf Bernhardt

Laboratory of Biophysics, Saarland University, Saarbrücken, Germany

During the last decade our group together with the groups of L. Kaestner and C. Wagner was able to demonstrate that red blood cells (RBCs) can play an active role in thrombus formation [1]. We were able to show that the enhancement of the intracellular Ca^{2+} content of RBCs induced by lysophosphatidic acid (LPA, substance released by activated thrombocytes) treatments results in the exposure of phosphatidylserine (PS) on the outer leaflet of the cell membrane due to the activation of the scramblase. In addition, it leads to cells shrinkage due to the activation of the Ca^{2+}-activated K^+ channel and the resulting KCl loss. Such a procedure could be observed for the majority of RBCs. However with confocal fluorescence microscopy and double labelling of the RBCs we observed that some cells show PS exposure without increased Ca^{2+} content and some other cells show enhanced Ca^{2+} content not resulting in significant PS exposure [2]. Experimental data and an explanation of such individual behaviour will be presented. In addition, we will present data showing a role of the Ca^{2+}-activated K^+ channel for the PS exposure. Finally, we will show that the observed effect is not depending on the age of the cells.

[1] Kaestner, L. et al. (2004) Thromb Haemost 92: 1269-72.

[2] Nguyen, D.B. et al. (2011) Cell Physiol Biochem 28: 847-856.

P.90 Polar protein localization

L. Wettmann, M. Bonny, and K. Kruse

Theoretical Physics, Saarland University, Saarbrücken, Germany

The distributions of many proteins in rod-shaped bacteria are far from homogeneous. Often they accumulate at the cell poles or in the cell center. At the same time, the copy number of proteins in a single cell is relatively small making the patterns noisy. To explore limits to protein patterns due to molecular noise, we studied a generic mechanism for spontaneous polar protein assemblies in rod-shaped bacteria, which is based on cooperative binding of proteins to the cytoplasmic membrane. For mono-polar assemblies, we find that the switching time between the two poles increases exponentially with the cell length and with the protein number. A symmetric protein distribution that is a stable state of the deterministic equations is not observed in the stochastic system.

P.91 Identifying transcription factor complexes and their roles

Will, Thorsten and Helms, Volkhard

Center for Bioinformatics, Saarland University, Saarbrücken, Germany

Eukaryotic gene expression is controlled through molecular logic circuits that combine regulatory signals of many different factors. In particular, complexation of transcription factors and other regulatory proteins is a prevailing and highly conserved mechanism of signal integration within critical regulatory pathways and enables us to infer controlled genes as well as the exerted regulatory mechanism. Common approaches for protein complex prediction that only use protein interaction networks, however, are designed to detect self-contained functional complexes and have difficulties to reveal dynamic combinatorial assemblies of physically interacting proteins.
We developed the novel algorithm DACO that combines protein-protein interaction networks and domain-domain interaction networks with

the cluster-quality metric cohesiveness. The metric is locally maximized on the holistic level of protein interactions and sophisticated connectivity constraints on the domain level are used to account for the exclusive and thus inherently combinatorial nature of the interactions within such assemblies.
When applied to predict transcription factor complexes in the yeast, the proposed approach outperformed popular complex prediction methods by far. Furthermore, we were able to assign many of the predictions to target genes, as well as to a potential regulatory effect in agreement with literature evidence.

P.92 Self-organization of acto-myosin leads to closure of the cytokinetic ring in mammalian cells

V. Wollrab[1,2], A. Wald[2], E. Wollrab[2], K. Kruse[2], D. Riveline[1]

[1] *Laboratory of Cell Physics, ISIS/IGBMC, Strasbourg, France*
[2] *Theoretical Physics, Saarland University, Saarbrücken, Germany*

Cytokinesis is the final step of cell division during which a cell is separated into two cells. 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 motors and filaments generates the stress necessary for contraction is still poorly understood. Here, we orient the cytokinetic ring in mammalian cells by placing individual cells in designed microcavities [1,2]. Our approach allows visualising the ring in a single plane of focus. We reveal a pattern of regular clusters of myosin and formin. These structures are stable and follow a radial trajectory throughout closure. In addition, formation of the clusters coincides with the onset of constriction. We propose that the myosin/formin pattern is self-organised and that its emergence is associated with a sharp increase in the stress generated by the acto-myosin ring. These results are supported by a continuum mean field model for active gels.

[1] Riveline, D. and Buguin, A. Devices and methods for observing the cell division. WO/2010/092116. *Patent* (2009).

[2] Riveline, D. and Wollrab, V. Devices and methods for observing eukaryotic cells without cell wall. WO/2013/135809. *Patent* (2012).

P.93 Increasing complexity out of coexisting autocatalysts

Emanuel Gregor Worst[1], Philipp Zimmer[2], Eva Wollrab[1], Karsten Kruse[2], Albrecht Ott[1]

[1]Biological Experimental Physics Department, [2]Theoretical Biological Physics Department, Saarland University, Saarbrücken, Germany

The evolution of an ecosystem in terms of the entity of all coexisting species is poorly understood. We examine a simplified system of auto-catalyzing, linear molecular chains that feed on each other. We ask how all of the species can robustly evolve towards a state of increased complexity as opposed to getting stuck by generating a single "winner". We present an experimental as well as a theoretical realization, generating continuously new configurations of species. We find that a selective formation of advanced configurations is easy to realize theoretically, while experimentally, it requires careful tuning of the parameters. This may well explain why, to our knowledge, a one dimensional molecular ecosystem did not emerge as a result of natural evolution.

P.94 Impact of bystander cells on cytotoxic efficiency of killer cells

Xiao Zhou[1], Mathieu Mangeat[2,3], Karsten Schwarz[2], Carsten Kummerow[1], Eva C. Schwarz[1], Cora Stephan[1], Heiko Rieger[2], Markus Hoth[1], and Bin Qu[1]

[1]*Department of Biophysics, School of Medicine, Saarland University, Homburg, Germany*

[2]*Theoretical Physics, Saarland University, Saarbrücken, Germany*

[3]*Laboratoire de Physique, Statistique, ENS, Paris, France*

Killer cells in the immune system, like cytotoxic T lymphocytes (CTL) and natural killer (NK) cells, protect the body by eliminating the pathogen infected cells and tumorigenic cells. The high specificity of this killing process to particular target cells is ensured by the formation of immunological synapse (IS). To find the right target cells, killer cells have to search in an environment where the target cells are hidden among healthy bystander cells. Though it is well studied how killer cells eliminate their targets, the impact of innocent healthy bystander cells on the cytotoxic efficiency of killer cells is unclear. Using the newly developed real time killing assay, we first checked whether bystander cells are killed by primary human CTL and NK cells. The results show that the bystander cells were not affected when killer cells eliminate their targets. In addition, we examined how the killing efficiency is affected by bystander cells. Certain numbers of target cells were mixed with bystander cells and killing efficiency was analyzed by the real-time killing assay. We found that with increasing number of bystander cells, the killing efficiency was slightly enhanced. We have established a model to simulate the behavior of killer cells with or without the presence of bystander cells. The preliminary results show that the presence of bystander cells can locally accelerate the migration of killer cells in contact with them and thereby increases the global killing efficiency. To confirm this prediction, we performed the same experiment on high content imaging system Bioimager to visualize the movement of killers, we found NKs migrated much faster with the presence of bystander cells than that without bystanders. Here we report for the first time, to our knowledge, that bystander cells can

enhance the killing efficiency of CTL and NKs and this impact might be resulted from the acceleration of killer searching velocity by bystander cells.

P.95 Evolution of increasingly complex linear molecules

Philipp Zimmer[1], Emanuel Gregor Worst[2], Eva Wollrab[2], Albrecht Ott[2], and Karsten Kruse[1]

[1]Theoretische Biologische Physik, [2]Biologische Experimentlphysik, Saarland University, Saarbrücken, Germany

Darwinian evolution is based on variation and selection acting on mutations, reproduction, or the metabolism of a species. These processes can only take place when the underlying system is out of thermodynamical equilibrium. For natural evolution the species as well as their relation network has continuously been gaining complexity. The conditions necessary for a steady increase in complexity are not well understood. Performing stochastic simulations as well as experiments with DNA, we analyze a chemical system consisting of autocatalytically concatenating chains. We find that, despite its inherent stochastic nature, the system evolves along a reproducible path towards states of increasing complexity if the autocatalytic activity exceeds a critical value.

P.96 Novel strategies to characterize Sec61-associated proteins regulating the transport of precursor proteins into the mammalian ER

Hoffmann, A., Ziska, A., Haßdenteufel, S., Schorr, S. and Zimmermann, R.

Department of Medical Biochemistry & Molecular Biology, Saarland University, Homburg, Germany

The transport of proteins into the mammalian endoplasmic reticulum (ER) is promoted by the dynamic Sec61 channel which is surrounded by a set of diverse accessory proteins. ER- specific signal peptides mediate the targeting of precursor protein to the ER membrane and facilitate the gating of the Sec61 translocon from the closed to the open state. Depending on the targeting pathway of a nascent polypeptide chain to the membrane-embedded Sec61, the import into the ER can occur either co- or posttranslational. However some precursors require for efficient translocation, besides an appropriate signal peptide, various of the Sec61-associated proteins. We are interested in determining the accessory proteins necessary for the co- and posttranslational transport of different polypeptides across the ER membrane, especially regarding the properties of the signal peptide. Model proteins were chosen regarding length and properties of their signal peptide as well as the total length of the mature protein. The influence of these ER proteins on targeting and translocation of different precursors was investigated by siRNA based depletion using an *in vitro* translation assay which involves semi-intact cells as well as intact living cells. Due to the natively structured ER membrane of human origin within a defined cytosolic fraction, semi-permeabilized cells were used to investigate exclusively the *in vitro* transport of one model protein. Moreover, to uncover the precursor of selected substrates and detect a translocation defect by its accumulation, we used plasmid-driven overexpression combined with a proteasome inhibitor in intact cells. This approach allows the detection of all protein forms synthesized by the cells including secreted proteins.

List of Participants

Ackermann, Björn	Saarland University, Saarbrücken, Germany *b.ackermann@physik.uni-saarland.de*
Akulenko, Ruslan	Saarland University, Saarbrücken, Germany *r.akulenko@bioinformatik.uni-saarland.de*
Alansary, Dalia	Saarland University, Homburg, Germany *ptdaan@uks.eu*
Alert, Ricard	University of Barcelona, Barcelona, Spain *ricardaz@ecm.ub.edu*
Amini, Mahnaz	Saarland University, Homburg, Germany *mah.amini4@gmail.com*
Angenendt, Adrian	Saarland University, Saarbrücken, Germany *adrian.angenendt@gmx.de*
Aouane, Othmane	Saarland University, Saarbrücken, Germany *o.aouane@mx.uni-saarland.de*
Appert-Rolland, Cecile	Univ. Paris Sud, Orsay, France *cecile.appert-rolland@th.u-psud.fr*
Arend, Judith	Saarland University, Saarbrücken, Germany *judith.arend@uks.eu*
Arita, Chikashi	Saarland University, Saarbrücken, Germany *c.arita@physik.uni-saarland.de*
Astanina, Ksenia	Saarland University, Saarbrücken, Germany *k.astanina@mx.uni-saarland.de*
Backes, Christian	Saarland University, Saarbrücken, Germany *christian.backes@uks.eu*
Bahr, Daniel	Saarland University, Saarbrücken, Germany *bahr@lusi.uni-sb.de*
Bandmann, Vera	INM Leibniz Institut for New Materials, Saarbrücken, Germany *vera.bandmann@inm-gmbh.de*
Barghash, Ahmad	Saarland University, Saarbrücken, Germany *barghash@bioinformatik.uni-saarland.de*
Bar-Ziv, Roy	Weizmann Institute of Science, Rehovot, Israel *Roy.Bar-Ziv@weizmann.ac.il*

Baumgart, Johannes	Max Planck Institute for the Physics of Complex Systems, Dresden, Germany *baumgart@pks.mpg.de*
Beck, Andreas	Saarland University, Saarbrücken, Germany *andreas.beck@uks.eu*
Becker, Björn	Saarland University, Saarbrücken, Germany *bjoern_beckr2@gmx.de*
Belkacemi, Thabet	Saarland University, Homburg, Germany *thabet.belkacemi@uks.eu*
Bermitt, Eric	Universität Bremen, Germany *hgd@uni-bremen.de*
Bischoff, Markus	Saarland University, Homburg, Germany *markus.bischoff@uks.eu*
Bleymehl, Katherin	Saarland University, Homburg, Germany katherin.bleymehl@gmx.de
Blum, Andreas	Saarland University, Saarbrücken, Germany *andreablum90@web.de*
Bogatikov, Evgenii	Saarland University, Homburg, Germany bogatikov@gmail.com
Bogeski, Ivan	Department of Biophysics, Homburg, Germany *ivan.bogeski@uks.eu*
Bonny, Mike	Saarland University, Saarbrücken, Germany *mike@lusi.uni-sb.de*
Brochard-Wyart,	Institut Curie-UPMC, Paris, France *brochard@curie.fr*
Bruns, Dieter	Saarland University, Homburg, Germany dieter.bruns@uks.eu
Casademunt, Jaume	Universitat de Barcelona, Barcelona, Spain *jaume.casademunt@ub.edu*
Cavalié, Adolfo	Saarland University, Homburg, Germany adolfo.cavalie@uks.eu
Chang, Hsin-Fang	Saarland University, Homburg, Germany *hsinfc@gmail.com*
Clavería, Viviana	Saarland University, Saarbrücken, Germany *viviana.claveria@usach.cl*
Colanesi, Sarah	Saarland University, Homburg, Germany *sarah.colanesi@uniklinikum-saarland.de*
Colette, Florimond	JKU Linz, Linz, Austria *florimond.collette@jku.at*
Cortassa, Sonia	John Hopkins University, Baltimore, USA

	scortas1@jhmi.edu
Dasgupta, Sabyasachi	Jülich Forschungszentrum, Jülich, Germany *s.dasgupta@fz-juelich.de*
de Jonge, Niels	INM Leibniz Institut for New Materials, Saarbrücken, Germany *niels.dejong@inm-gmbh.de*
Deek, Joanna	Technische Universität München, Garching bei München, Germany *joanna.deek@tum.de*
Dembla, Ekta	Saarland University, Homburg, Germany *ekta.dembla@uniklinikum-saarland.de*
den Otter, Wouter	University of Twente, Enschede, The Netherlands *w.k.denotter@utwente.nl*
Dhara, Madhurima	Saarland University, Homburg, Germany *madhurimadhara@gmail.com*
Döbereiner, Hans-Günther	Universität Bremen, Bremen, Germany *hgd@uni-bremen.de*
Dobicki, Heike	Saarland University, Saarbrücken, Germany *heike.dobicki@physik.uni-saarland.de*
Dreher, Alexander	Saarland University, Saarbrücken, Germany *adreher@lusi.uni-sb.de*
Eckrich, Tobias	Saarland University, Homburg, Germany *t.eckrich@mx.uni-saarland.de*
Elgeti, Jens	Jülich Forschungszentrum, Jülich, Germany *j.elgeti@fz-juelich.de*
Engel, Jutta	Saarland University, Homburg, Germany *jutta.engel@mx.uni-saarland.de*
Erlenkämper, Christoph	Institut Curie, Paris, France *christoph.erlemkaemper@curie.fr*
Erol, Arzu	Saarland University, Homburg, Germany *arzubaharerol@yahoo.com*
Faidt, Thomas	Saarland University, Saarbrücken, Germany *t.faid@physik.uni-saarland.de*
Feng, Jin	Saarland University, Homburg, Germany *jin.jane.feng@gmail.com*
Flesch, Julia	Saarland University, Saarbrücken, Germany *julflesch@aol.com*
Fleury, Jean Baptiste	Saarland University, Saarbrücken, Germany *jean-baptiste.fleury@physik.uni-saarland.de*

Flormann, Daniel	AG-Wagner, Saarbrücken, Germany *daniel.flormann@online.de*
Foffano, Giulia	LPTMS- Université Paris-Sud, Paris, France *giulia.foffano@u-psud.fr*
Friedrich, Thierry	Saarland University, Saarbrücken, Germany *thierry@lusi.uni-sb.de*
Frolov, Vadim	University of Basque Country, Leioa, Spain *vadim.frolov@ehu.es*
Gadelha, Hermes	University of Oxford, Oxford, United Kingdom *gadelha@maths.ox.ac.uk*
Gamayun, Igor	Saarland University, Homburg, Germany *igor.gamayun@uks.eu*
Gartmann, Julia	Saarland University, Homburg, Germany *julia.gartmann@freenet.de*
Giehr, Pascal	Saarland University, Saarbrücken, Germany *pascalgiehr@googlemail.com*
Goswami, Debajit	Saarland University, Saarbrücken, Germany *debajit@lusi.uni-sb.de*
Grass, Simone	Saarland University, Homburg, Germany *Simone.grass@uniklinikum-saarland.de*
Gratz, Micha	Saarland University, Saarbrücken, Germany *micha.gratz@uni-saarland.de*
Gross, Steven	UC Irvine, Irvine, USA *grosslab@gmail.com*
Haefner, Sabrina	Saarland University, Saarbrücken, Germany *s.haefner@physik.uni-saarland.de*
Hafner, Anne	Saarland University, Saarbrücken, Germany anne.hafner@lusi.uni-sb.de
Hähl, Hendrik	Saarland University, Saarbrücken, Germany *h.haehl@physik.uni-saarland.de*
Hakim, Vincent	Ecole Normale Supérieure, Paris, France *vincent.hakim@ens.fr*
Hamed, Mohamed	Saarland University, Saarbrücken, Germany mhamed@bioinformatik
Hannig, Matthias	Saarland University, Homburg, Germany *matthias.hannig@uks.eu*
Harb, Ali	Saarland University, Homburg, Germany ali.harb@uks.eu
Hein, Michael	Saarland University, Saarbrücken, Germany *michael.hein@physik.uni-saarland.de*

Helms, Volkhard	Saarland University, Saarbrücken, Germany *volkhard.helms@bioinformatik.uni-saarland.de*
Herrmann, Mathias	Saarland University, Homburg, Germany *mathias.hermann@uks.eu*
Hoffmann, Alexandra	Saarland University, Homburg, Germany *hoffmann.a@saarhorst.de*
Hoth, Markus	Saarland University, Saarbrücken, Germany *markus.hoth@uks.eu*
Hudalla, Philipp	Saarland University, Saarbrücken, Germany p.hudalla@lusi.uni-sb.de
Hufnagel, Lars	European Molecular Biology Laboratory, Heidelberg, Germany *hufnagel@embl.de*
Hui, Xin	Saarland University, Homburg, Germany *xin.hui@uks.eu*
Inamdar, Mandar	Indian Institute of Technology Bombay, *Mumbai, India mandarinamdar78@gmail.com*
Jacobs, Karin	Saarland University, Saarbrücken, Germany *k.jacobs@physik.uni-saarland.de*
Janmey, Paul	University of Pennsylvania, Philadelphia, USA *janmey@mail.med.upenn.edu*
Joanny, Jean-François	ESPCI, Paris, France *jean-francois.joanny@espci.fr*
Johann, Denis	Saarland University, Saarbrücken, Germany *johann@lusi.uni-sb.de*
Jourdain, Dominic	Saarland University, Saarbrücken, Germany *domi@lusi.uni-sb.de*
Jülicher, Frank	Max Planck Institute PKS, Dresden, Germany *julicher@pks.mpg.de*
Jung, Philipp	Saarland University, Homburg, Germany *philipp.jung@uks.eu*
Kainka, Lucina	Saarland University, Saarbrücken, Germany *l.kainka@web.de*
Kaiser, Elisabeth	Saarland University, Homburg, Germany *elisabeth.kaiser@uks.eu*
Kappel, Sven	Saarland University, Homburg, Germany *sven.kappel@uks.eu*
Karsch, Susanne	Georg-August University, Goettingen, Germany *susanne.karsch@phys.uni-goettingen.de*
Käs, Josef	University of Leipzig, Leipzig, Germany

	lkaes@physik.uni-leipzig.de
Kierfeld, Jan	TU Dortmund, Dortmund, Germany *jan.kierfeld@tu-dortmund.de*
Kihm, Alexander	Saarland University, Saarbrücken, Germany *alexanderkihm@gmail.com*
Kircher, Sarah	Saarland University, Homburg, Germany *sarah.kircher@uniklinikum-saarland.de*
Klein, Sarah	Saarland University, Saarbrücken, Germany *sklein@lusi.uni-sb.de*
Klos, Mischa	Saarland University, Saarbrücken, Germany *m.klos@physik.uni-saarland.de*
Klumpp, Stefan	Max Planck Institute of Colloids and Interfaces, Potsdam, Germany *klumpp@mpikg.mpq.de*
Knörk, Arne	Saarland University, Saarbrücken, Germany *s9arknoe@stud.uni-saarland.de*
Konrad, Maik	Saarland University, Saarbrücken, Germany *maik.konrad@uks.eu*
Kreis, Christian	Saarland University, Saarbrücken, Germany *c.kreis@physik.uni-saarland.de*
Kreysing, Moritz	Max Planck Institute for Cell Biology, Dresden, Germany *kreysing@mpi-cbg.de*
Kruse, Kartsten	Saarland University, Saarbrücken, Germany *k.kruse@physik.uni-sarland.de*
Kummerow, Carsten	Saarland University, Homburg, Germany *carsten.kummerow@uks.eu*
Kyriakopoulos, Charalampos	Saarland University, Saarbrücken, Germany *s9chkyri@stud.uni-saarland.de*
Lancaster, C. Roy D.	Saarland University, Homburg, Germany *roy.lancaster@structural-biology.eu*
Lannoo, Bruno	KU Leuven, Leuven, Belgium *brunolannoo@gmail.com*
Lautenschläger, Franziska	Saarland University, Saarbrücken, Germany *f.lautenschlaeger@physik.uni-saarland.de*
Lee, Po-Hsien	Saarland University, Saarbrücken, Germany *p.lee@mx.uni-saarland.de*
Leidinger, Olaf	Saarland University, Saarbrücken, Germany *oleid@lusi.uni-sb.de*

Leschik, Susanne	DGFB, Halberstadt, Germany *masteringenieur@gmail.com*
Leschik, Holger	DGFB, Halberstadt, Germany *masteringenieur@gmail.com*
Lewis, Richard	Stanford University, Stanford, USA *rslewis@stanford.edu*
Li, Menglin	Saarland University, Saarbrücken, Germany *leonard.li@uni-saarland.de*
Li, Qian	Saarland University, Homburg, Germany *sleepingbaby81@hotmail.com*
Lipowsky, Reinhard	Max Planck Institute of Colloids and Interfaces, Potsdam, Germany *lipowsky@mpikg.mpg.de*
Lis, Annette	Saarland University, Homburg, Germany *annette.lis@uniklinikum-saarland.de*
Liu, Jian	NIH, Bethesa, USA *jian.liu@nih.gov*
Loehfelm, Aline	Saarland University, Saarbrücken, Germany *s9alloeh@stud.uni-saarland.de*
Lück, Alexander	Saarland University, Saarbrücken, Germany *alexander.lueck@lusi.uni-sb.de*
Lyrmann, Helene	Saarland University, Saarbrücken, Germany *helene.lyrmann@uks.eu*
Maccari, Ilaria	Saarland University, Saarbrücken, Germany *ilariamaccari@libero.it*
Maitra, Ananyo	Indian Institute of Science, Bangalore, India *nyomaitra07@gmail.com*
Mamane, Alexandre	Institut Curie, Paris, France *alexandre.mamane@curie.fr*
Mang, Sebastian	Saarland University, Homburg, Germany *sebastian.mang@t-online.de*
Marganska, Magdalena	University of Regensburg, Germany *magdalena.marganska@physik.uni-regensburg.de*
McKenney, Richard	University of California, San Francisco, USA *richard.mckenney@ucsf.edu*
Miederer, Anna-Maria	Saarland University, Homburg, Germany *anna-maria.miederer@uks.eu*
Minc, Nicolas	Institut Jacques Monod, Paris, France *minc@ijm.univ-paris-diderot.fr*

Mittasch, Matthäus	Technische Universität Dresden, Dresden, Germany *matthaeus.mittasch@mpi-cbg.de*
Mohammadi, Mina	Saarland University, Saarbrücken, Germany *m.mohammadi@physik.uni-saarland.de*
Mohrmann, Ralf	Saarland University, Homburg, Germany *ralf.mohrmann@uks.eu*
Monzon, Gina	Saarland University, Saarbrücken, Germany *g.monzon@lusi.uni-sb.de*
Müller, Frank	Saarland University, Saarbrücken, Germany *f.mueller@mx.uni-saarland.de*
Münkner, Stefan	Saarland University, Homburg, Germany *stefan.muenkner@mx.uni-saarland.de*
Najafi, Javad	Saarland University, Saarbrücken, Germany *jnajafi.phys@gmail.com*
Nazarieh, Maryam	Saarland University, Saarbrücken, Germany *maryam.nazarieh@gmail.com*
Needleman, Daniel	Harvard, Cambridge, USA *dan.needleman@gmail.com*
Neef, Marc	Saarland University, Saarbrücken, Germany *marcneef@lusi.uni-sb.de*
Nguyen, Duy	Saarland University, Saarbrücken, Germany *duy.nguyen@bioinfomatik.uni-saarland.de*
Nicolas, Thewes	Saarland University, Saarbrücken, Germany *n.thewes@physik.uni-saarland.de*
Niemeyer, Barbara	Saarland University, Homburg, Germany *barbara.niemeyer@uks.eu*
Nirody, Jasmin	University of California, Berkeley CA, USA *jnirody@berkeley.edu*
Nitze, Katja	Saarland University, Saarbrücken, Germany *katja.nitze@uks.eu*
Nosal, Olena	International Center for Molecular Physiology NASU, Kyiv, Ukraine *elena_n@biph.kiev.ua*
Okello, Emmanuel	Kampala University, Kampala, Uganda *emmens88@yahoo.com*
Onuchic, Jose	Rice University, Houston TX, USA *jonuchic@rice.edu*
Oriola, David	University of Barcelona, Barcelona, Spain *oriole@ecm.ub.edu*

Paknikar, Aishwarya	Istitute for X-Ray Physics, Göttingen, Germany *aishwarya.paknikar@phys.uni-goettingen.de*
Parker, Ian	University of California, Irvine CA, USA *iparker@uci.edu*
Pasula, Aravind	Saarland University, Saarbrücken, Germany *a.pasula@mx.uni-saarland.de*
Peckys, Diana	INM Leibniz Institut for New Materials, Saarbrücken, Germany *diana.pecky@inm-gmbh.de*
Peglow, Martin	Saarland University, Saarbrücken, Germany *martinpeglow@gmx.de*
Peinelt, Christine	Saarland University, Homburg, Germany *bpcpei@uks.eu*
Peisker, Henrik	Saarland University, Homburg, Germany *henrik.peisker@uks.eu*
Petrasek, Zdenek	MPI f. Biochemistry, Martinsried, Germany *petrasek@biochem.mpg.de*
Pick, Tillman	Saarland University, Homburg, Germany *tillman.pick@uniklinikum-saarland.de*
Poppenborg, Friederike	Saarland University, Saarbrücken, Germany *s9frpopp@stud.uni-saarland.de*
Prost, Jacques	Institut Curie, Paris, France *jacques.prost@curie.fr*
Qu, Bin	Saarland University, Homburg, Germany *bin.qu@uks.eu*
Rammo, Domenik	Saarland University, Saarbrücken, Germany doramm@gmx.de
Rehage, Benjamin	Saarland University, Homburg, Germany *benjamin.rehage@uks.eu*
Rehage, Melanie	Saarland University, Homburg, Germany *melanie.rehage@uks.eu*
Rieger, Heiko	Saarland University, Saarbrücken, Germany *h.rieger@mx.uni-saarland.de*
Risler, Thomas	Institut Curie, Paris, France *thomas.risler@curie.fr*
Riveline, Daniel	Université de Strasbourg, Strasbourg, France *riveline@unistra.fr*
Rückel, Martin	Humbold University Berlin, Berlin, Germany *mrueckl@physik.hu-berlin.de*
Rüdinger, Sten	Humbold University Berlin, Berlin, Germany *sten.ruedinger@physik.hu-berlin.de*

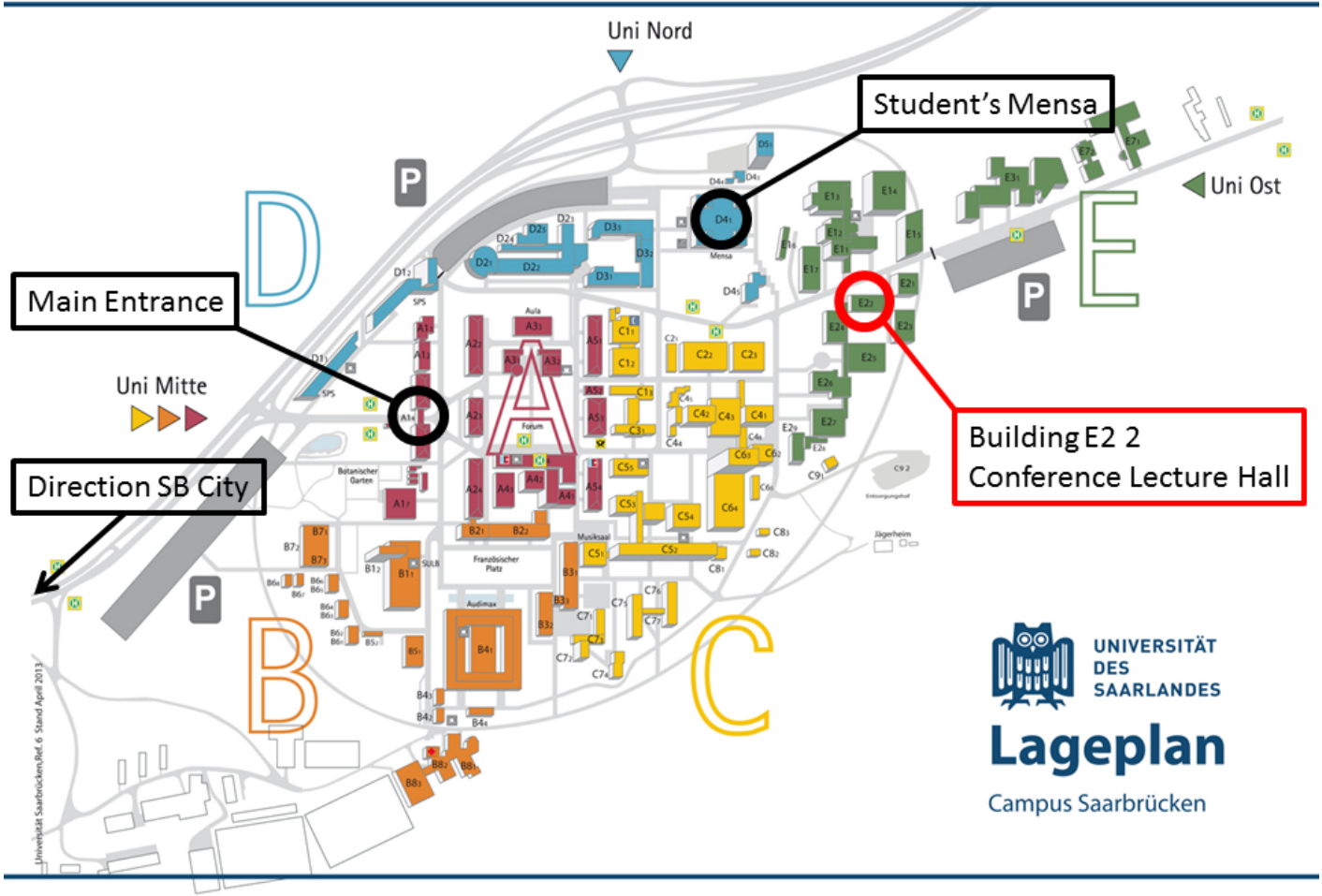
Rupf, Stefan	Saarland University, Homburg, Germany *stefan.rupf@uks.eu*
Sabass, Benedikt	Princeton University, Princeton NJ, USA *benedikt.sabass@gmx.de*
Sadjadi, Zeinab	Saarland University, Saarbrücken, Germany *sadjadi@lusi.uni-sb.de*
Sain, Anirban	Indian Institute of Technology- Bombay, Mumbai, India *asain@phy.iitb.ac.i*
Salbreux, Guillaume	Max Planck Institute for the Physics of Complex Systems, Dresden, Germany *salbreux@pks.mpg.de*
Sander, Mathias	Saarland University, Saarbrücken, Germany *m.sander@physik.uni-saarland.de*
Santen, Ludger	Saarland University, Saarbrücken, Germany *santen@lusi.uni-sb.de*
Saul, Stephanie	Saarland University, Saarbrücken, Germany *stephanie.saul@uks.eu*
Schenkelberger, Marc	Saarland University, Saarbrücken, Germany *marcschenkelberger@t-online.de*
Scherer, Sabrina	Saarland University, Saarbrücken, Germany *sabrina.scherer@physik.uni-saarland.de*
Schlenstedt, Gabriel	Saarland University, Homburg, Germany *gabriel.schlenstedt@uniklinikum-saarland.de*
Schmitt, Manfred	Saarland University, Saarbrücken, Germany *mjs@microbiol.uni-sb.de*
Schoppmeyer, Rouven CD	Saarland University, Homburg, Germany *r.schoppmeyer@gmail.com*
Schorr, Stefan	Saarland University, Homburg, Germany *s9stsch@stud.uni-saarland.de*
Schröder, Laura	Saarland University, Homburg, Germany *laura.schroeder@uniklinikum-saarland.de*
Schwarz, Karin	Saarland University, Homburg, Germany *dr.karin.schwarz@uks.eu*
Schwarz, Eva	Saarland University, Homburg, Germany *eva.schwarz@uks.eu*
Schwarz, Karsten	Saarland University, Saarbrücken, Germany *corsa@lusi.uni-sb.de*
Seemann, Ralf	Saarland University, Saarbrücken, Germany *r.seemann@physik.uni-saarland.de*

Serwe, Timo	Saarland University, Saarbrücken, Germany timo.serwe@lusi.uni-sb.de
Shaebani, Reza	Saarland University, Saarbrücken, Germany *shaebnai@lusi.uni-sb.de*
Shaib, Ali	Saarland University, Homburg, Germany *ali.shaib@uks.eu*
Soumya, S S	Indian Institute of Technology Bombay, India *soumyass@iitb.ac.in*
Spengler, Christian	Saarland University, Saarbrücken, Germany *c.spengler@physik.uni-saarland.de*
Stankevicins, Luiza	Saarland University, Saarbrücken, Germany *l.stankevicins@physik.uni-saarland.de*
Stentenbach, Gabriele	Saarland University, Saarbrücken, Germany *g.stentenbach@physik.uni-saarland.de*
Suiwal, Shweta	Saarland University, Homburg, Germany *shwetasuiwal@gmail.com*
Tamiello, Chiara	Eindhoven Univ. of Technology, Netherlands *c.tamiello@tue.nl*
Tanimoto, Hirokazu	Institut Jacques Monod CNRS, Paris, France *tanimoto.hirokazu@ijm.univ-paris-diderot.fr*
Tarle, Victoria	Weizmann Institute of Science, Rehovot, Israel *tarle.victoria@weizmann.ac.il*
Terriac, Emmanuel	Saarland University, Saarbrücken, Germany *emmanuel.terriac@physik.uni-saarland.de*
Thewes, Alexander	Saarland University, Saarbrücken, Germany *alexth@lusi.uni-sb.de*
Thewes, Nicolas	Saarland University, Saarbrücken, Germany *n.thewes@physik.uni-saarland.de*
Thome, Christian	Saarland University, Saarbrücken, Germany *thome@lusi.uni-sb.de*
Tinschert, René	Saarland University, Homburg, Germany *rene.tinschert@gmail.com*
Turlier, Hervé	EMBL, Heidelberg, Germany *herve.turlier@embl.de*
Ullrich, Alexander	Freie Universität Berlin, Berlin, Germany *alexander.ullrich@fu-berlin.de*
Umanskaya, Natalia	Saarland University, Homburg, Germany *natalia.umanskaya@uks.eu*
Vandanapu, RamaRamesh	Saarland University, Homburg, Germany *ramaramesh113@gmail.com*

Vargas, Nabor	Saarland University, Saarbrücken, Germany *nabor.vargas@physik.uni-saarland.de*
Visco, Paolo	Université Paris-Diderot, Paris, France *paolo.visco@univ-paris-diderot.fr*
Vishavkarma, Renu	Raman Research Institute, Bangalore, India *renuv@rri.res.in*
Wagner, Christian	Saarland University, Saarbrücken, Germany *c.wagner@mx.uni-saarland.de*
Walter, Jörn	Saarland University, Saarbrücken, Germany *j.walter@mx.uni-saarland.de*
Weber, Tobias	Saarland University, Saarbrücken, Germany *tob.weber90@gmail.com*
Weber, Ines	Saarland University, Saarbrücken, Germany *weber@lusi.uni-sb.de*
Wedlich-Söldner, Roland	University of Münster, Münster, Germany *wedlich@uni-muenster.de*
Welter, Michael	Saarland University, Saarbrücken, Germany *mwelter@lusi.uni-sb.de*
Wesseling, Mauro Carlos	Saarland University, Saarbrücken, Germany *m.wesseling@mx.uni-saarland.de*
Wettmann, Lukas	Saarland University, Saarbrücken, Germany *lwettmann@gmx.de*
Will, Thorsten	Saarland University, Saarbrücken, Germany *thorsten.will@bioinformatik.uni-saarland.de*
Wolf, Verena	Saarland University, Saarbrücken, Germany *wolf@cs.uni-saarland.de*
Wollrab, Eva	Saarland University, Saarbrücken, Germany *evawollrab@gmx.de*
Wollrab, Viktoria	Saarland University/Strasbourg, Strasbourg, France *wollrab@unistra.fr*
Worst, Emanuel	Saarland University, Saarbrücken, Germany *m.worst@physik.uni-saarland.de*
Xin, Zhang	Saarland University, Homburg, Germany *zhangxin02st@gmail.com*
Yarzagaray, Antonio	Saarland University, Saarbrücken, Germany *antonio.yarzagaray@hotmail.com*
Yule, David	University of Rochester, Rochester PA, USA *david_yule@urmc.rochester.edu*

Zhao, Renping	Saarland University, Homburg, Germany *renping.zhao@gmail.com*
Zhou, Xiao	Saarland University, Homburg, Germany *xiao.zhou@uks.eu*
Zhou, Yan	Saarland University, Homburg, Germany *yvonnezyjj@gmail.com*
Ziebert, Falko	Albert-Ludwigs-Universität Freiburg, Freiburg, Germany *fziebert@gmail.com*
Zimmer, Philipp	Saarland University, Saarbrücken, Germany *philipp@lusi.uni-sb.de*
Zimmermann, Katharina	Saarland University, Homburg, Germany *katharina.zimmermann@uniklinikum-saarland.de*
Ziska, Anke	Saarland University, Homburg, Germany *anke.ziska@gmx.de*

Author Index



http://www.uni-saarland.de/footer/dialog/anfahrt/lageplan/interaktiv.html

	Monday (22.9.)	Tuesday (23.9)	Wednesd.(24.9)	Thursd. (25.9.)	Friday (26.9.)
8:50-9:00		Conf.-Opening			
9:00-9:45		J. Prost	J. Käs	P. Janmey	F. Brochard
9:45-10:30		R. McKenney	N. Minc	L. Hufnagel	J.-F. Joanny
10:30-11:00		Coffee break	Coffee break	Coffee break	Coffee break
11:00-11:30		J. Liu	J. Casademunt	T. Risler	H.G. Döbereiner
11:30-12:00		D. Riveline	F. Ziebert	C. Erlenkämper	Z. Petrášek
12:00-12:30		K. Kruse	J. Kierfeld	S. Klumpp	M. Kreysing
12:30-14:00		Lunch	Lunch	Lunch	Closing / Lunch
14:00-14:45		S. Gross	D. Needleman	I. Parker	
14:45-15:30		R. Lipowsky	F. Jülicher	S. Cortassa	
15:30-16:00		Coffee break	Coffee break	Coffee break	
16:00-16:45		R. Lewis	V. Hakim	R. Bar-Ziv	
16:45-17:30		D. Yule	R. Wedlich-Söldner	J. Onuchic	
17:30-18:00		M. Hoth	C. Appert	J. Elgeti	
18:00-18:30		Poster-Session & Snacks			
18:30 –			Barbecue	Speaker Dinner	

Pre-Conference Workshop
(participation of externals free)