BP 3: Membranes and Interfaces

Time: Monday 14:00–17:15 Location: H43

Invited Talk BP 3.1 Mon 14:00 H43

On the Timescales of Membrane Fusion — •Rumiana Dimova — Max Planck Institute of Colloids and Interfaces, Science Park Golm, 14424 Potsdam, Germany

Membrane fusion is a vital process as it is involved in many cellular functions and stages of cell life like import of foodstuffs and export of waste, signaling between nerve cells, fertilization, and virus infection. In both the life sciences and bioengineering, controlled membrane fusion has many possible applications, such as drug delivery, gene transfer, chemical microreactors, or synthesis of nanomaterials.

Fusion dynamics is intriguing but microscopy observations with time resolution higher than several milliseconds have not been achieved until now. Using micromanipulation of giant unilamellar vesicles as model membranes one can directly observe membrane fusion. We induce the fusion of giant lipid vesicles in a controlled manner and monitor the fusion dynamics with a temporal resolution of 50 microseconds; see Haluska et al. Proc. Natl. Acad. Sci. USA. 103, 15841-15846 (2006). Two different approaches of inducing directed fusion are used: i) employing synthetic fusogenic molecules incorporated in the membranes, and ii) electrofusion. For both protocols, the opening of the fusion necks is very fast, with an average expansion velocity of centimeters per second. This velocity indicates that the initial formation of a single fusion neck can be completed in a few hundred nanoseconds.

BP 3.2 Mon 14:30 H43

Energy barriers for membrane fusion — •Andrea Grafmueller and Reinhard Lipowsky — Max-Planck-Institut of Colloids and Interfaces, Potsdam, Germany

The fusion of bilayer membranes and vesicles has been studied using Dissipative Particle Dynamics (DPD) simulations. A large number of fusion attempts between a vesicle and a planar membrane segment is monitored varying the area per lipid molecule which determines the initial membrane tension. Fusion events are observed with a high success rates at high tensions. For these successful events, the fusion time, i.e., the time from first contact between the bilayers until the opening of the pore, shows a strong, exponential dependence on the membrane tension. The observed fusion process starts with the adhesion of the vesicle to the tense planar segment. Inter-bilayer flipflops disturb the bilayer order near the rim of the adhered area. Finally, the molecules in this disordered region reorganize into a small segment of a single (hemifused) bilayer, which ruptures at the edge. A detailed analysis of the observed fusion events reveals that these events are governed by at least two successive energy barriers.

BP 3.3 Mon 14:45 H43

What can be learned from a coarse-grained description of membrane fusion? — •Marcus Muller — Institut fuer Theoretische Physik, Georg-August Universitaet, Goettingen

Membrane fusion is a fundamental biological process of importance in fertilization, synaptic release, intracellular traffic, and viral infection. Coarse-grained models can contribute to our understanding of these collective phenomena in membranes [1] that evolve on a few nanometers and milliseconds.

We have carried out simulations of the fusion of tense apposed bilayers formed by amphiphilic molecules. The fusion pathway differs from the common stalk mechanism. Stalks do form between the apposed bilayers, but rather than expand radially to form an axial-symmetric hemifusion diaphragm and they promote the nucleation of small holes in their vicinity. Then, the stalk encircles a hole in one bilayer creating a diaphragm which ruptures to complete the fusion pore. The pathway give rise to mixing between both leaves of the bilayer and allow for transient leakage.

Self-consistent field calculations have be used to explored the role of lipid architecture and tension, and to calculate free energy barriers along the fusion path. We find that (i) successful fusion is found to be severely limited by the architecture of the lipids and that (ii) any mechanism which affects even modestly the line tension of a hole in a membrane affects greatly the ability of that membrane to undergo fusion.

[1] M. Müller, K. Katsov, and M. Schick, Phys. Rep. 434, 113 (2006)

BP 3.4 Mon 15:00 H43

Lipid Nanotubes for Probing Cell Membrane Reservoir — •Darius V. Köster[1], Pierre Sens[2], Christophe Lamaze[1], and Pierre Nassoy[1] — [1]Institut Curie, Paris, France — [2]ESPCI, Paris, France

Cells are exposed to mechanical stress due to shear flow (e.g. in veins and arteries) or stretching and relaxation (e.g. in muscle tissue). In this study, we study the mechanisms, which provide membrane integrity during these processes, since the membrane as a pure lipid bilayer would be fairly inextensible, and any stretching of it would lead to rupture. One important parameter to describe the cell membrane is its membrane tension, and it is reported that cells have membrane reservoirs, and regulate membrane tension. Pulling small tubes out of the cell membrane in using an optical trap allows us to probe these reservoirs and to measure the membrane tension. In combination with biological tools of cell modification (transfection and drug treatment) and fluorescence imaging we aim at identifying the compartments involved in membrane tension regulation. More specifically, in this work, we will focus on the role of caveolae, which are small membrane invaginations, in membrane tension buffering. To get a clear picture of their mechanical function, we will show that the interaction between membrane and cytoskeleton has to be investigated in details. Finally, we will propose that caveolae can indeed act as available membrane reservoirs for a cell membrane to accommodate sudden extend stress.

BP 3.5 Mon 15:15 H43

A novel method for measuring the bending rigidity of model lipid membranes by simulating tethers — •Vagelis Harmandaris and Markus Deserno — Max-Planck-Institute for Polymer Research*Max-Planck-Institute for Polymer Research, Theory Group, Mainz, Germany

The most common approach for measuring bending rigidities in simulations is from the spectrum of thermal shape fluctuations, which is the analogous of the experimental *flicker spectroscopy* technique. An alternative experimental method is to measure the tensile force needed to pull nanoscale bilayer tubes (tethers) from vesicles, since this force is proportional to the membrane's bending modulus and inversely proportional to the tube radius. Here, we show that this relation can be applied with even greater ease in computer simulations. Using a coarse-grained bilayer model developed recently [1], we efficiently obtain bending rigidities that compare very well with complementary measurements based on an analysis of thermal undulation modes. We furthermore illustrate that no deviations from simple quadratic continuum theory occur up to a radius of curvature comparable to the bilayer thickness [2].

References: 1. I.R. Cooke, K. Kremer and M. Deserno, Phys. Rev. E 72, 011506 (2005). 2. V. Harmandaris and M. Deserno, J. Chem. Phys. 125, 204905 (2006).

BP 3.6 Mon 15:30 H43

Shape and fluctuations of biphasic membrane vesicles — •Stefan Semrau[1], Timon Idema[2], Cornelis Storm[2], and Thomas Schmidt[1] — [1]Physics of life processes, Leiden institute of physics, Leiden university, The Netherlands — [2]Theoretical biophysics, Lorentz institute, Leiden university, The Netherlands

Heterogeneities in the cell membrane due to coexisting lipid phases have been conjectured to play a major functional role in cell signaling and traffic. Purely physical properties of such multiphase systems, such as the line tension and the bending moduli, are crucially involved in endocytocis and lipid trafficking, and determine the kinetics and asymptotics of phase separation. We have developed an analytical description of the vesicle shape of weakly budded biphasic vesicles and shown it to be in excellent agreement with numerical calculations and experiments. Our description allows for a reproducible and reliable systematic determination of the physical parameters of the membrane in the biologically relevant limit of weakly budded shapes. The parameters thus obtained allow us to determine an upper bound for the size of nanodomains in the plasma membrane of living cells.

BP 3.7 Mon 15:45 H43

Structure and dynamics of crystalline protein layers peripherally bound to supported lipid bilayers — •Christian Reich, Margaret Horton, Joachim Rädler, and Bert Nickel — Depart-

ment für Physik, Ludwig-Maximilians-Universität, D-80539 München

We model peripheral membrane proteins at the surface of cell membranes using streptavidin and avidin bound to biotinylated lipids in a supported lipid bilayer (SLB) at the solid-liquid interface. Using X-ray reflectivity and simultaneous fluorescence microscopy, we characterize the structure and fluidity of a protein layer containing two-dimensional streptavidin crystals bound to a SLB. A single lipid bilayer provides a biologically-relevant environment for in-situ investigation of membrane-associated proteins interacting with lipids. Using continuous bleaching, we measure a 10-15% decrease in the fluidity of the SLB after protein layer formation. We propose that this reduction in lipid mobility is due to a small fraction ca. 0.04 of immobilized lipids bound to the protein layer that create obstacles to membrane diffusion. Fits to our X-ray reflectivity data show a ca. 40 Å thick layer of protein and we resolve the ca. 8 Å layer separating the protein layer from the bilayer. We suggest that the separation provided by this water layer allows the underlying lipid bilayer to retain its fluidity and stability. Finally, we show how complementary information can be obtained in neutron experiments at REFSANS (FRM2).

BP 3.8 Mon 16:00 H43

Curvature-mediated interactions between membrane proteins lead to aggregation and vesiculation — Benedict Reynolds, Gregoria Illya, Vagelis Harmandaris, Martin Müller, Kurt Kremer, and •Markus Deserno — MPI für Polymerforschung, Mainz, Germany

Cellular tasks such as endocytosis, vesiculation, and protein sorting, or the biogenesis of organelles such as the endoplasmic reticulum or the Golgi apparatus rely on significant protein-assisted membrane remodeling. Special curvature-sensitive proteins may both *experience* geometry-driven forces and, conversely, *induce* major changes in membrane shape and topology. But due to the lipid bilayer's bending stiffness, the latter requires the cooperative action of many individual proteins. The necessary protein aggregation is thought to be driven by specific interactions, but more generic mechanisms such as membrane mediated interactions are recently being discussed by biologists. I will show that the underlying physics of curvature forces is not as straightforward as it is sometimes assumed. Using large-scale coarse-grained membrane simulations I then demonstrate that even in the absence of direct protein interactions curvature-mediated forces alone provide a robust mechanism for aggregation and can subsequently trigger vesiculation.

BP 3.9 Mon 16:15 H43

Interplay of lateral diffusion and membrane fluctuations — •Ellen Reister-Gottfried and Udo Seifert — II. Institut für Theoretische Physik, Universität Stuttgart, 70550 Stuttgart, Germany

Using a simulation scheme that numerically integrates both the equation of motion of a membrane and the Langevin equation of a particle diffusing freely along the curved surface of the membrane we study the interplay of membrane fluctuations and lateral diffusion. The energy of the membrane is given by the Helfrich Hamiltonian and its shape in the Monge gauge. In the regime where the relaxation time of membrane undulations with wavelength ξ is much smaller than the average time it takes a particle to cover the distance ξ, the particle experiences only averaged membrane quantities, such that a preaveraging approximation can be employed. We compare the diffusion coefficient projected on a flat reference plane –this is the typically measured quantity– obtained in previous analytical calculations that make use of this approximation with simulation results. Although the simulation scheme overcomes preaveraging, there is a surprisingly good agreement of analytical and simulation results even for parameter sets that do not meet the conditions for the preaveraging approximation. A detailed analysis of appropriate correlation functions using the simulation scheme explains the large validity range of the approximation.

BP 3.10 Mon 16:30 H43

Diffusion of nano-particles in model membranes — •Florian Rückerl, Carsten Selle, and Josef Käs — Universität Leipzig, Institut für Experimentelle Physik I, Abt. PWM

Langmuir monolayers are used as a simple membrane model in which partially charged nano-particles diffuse as model proteins. This system provides good control over obstacle sizes. The condensed domains within liquid phases that are found in the coexistence region exhibit a net dipole moment. The radial dependence of this electric dipolar field changes with the size of the domains from $E(|\mathbf{r}|) \propto 1/|\mathbf{r}|^3$ for a single dipole to $E(|\mathbf{r}|) \propto 1/|\mathbf{r}|$ for large domains ($R > 10\mu m$). The influence of this change on the particle diffusion was investigated by Monte Carlo simulations. The analysis shows that the particles are stronger confined at the domain border of smaller domains and that a change from two to one dimensional diffusion occurs.

We further investigate a more complex system, nano-particles diffusing on the surface of giant unilamellar vesicles composed of either a single lipid or a mixture of lipids: DOPC, DPPC and cholesterol. The latter systems exhibit $L_d - L_o$ coexisting phases which were shown to form curvature gradients in their bilayer surfaces. Therefore, the influence of the local membrane curvature on the diffusive behavior of the nano-particles can be investigated. A variety of lipid compositions and particles, $R = 34nm - 1.6\mu m$ with varying surface modifications, are used in order to to elucidate the interactions between nano-particles and lipids in bilayer membranes.

BP 3.11 Mon 16:45 H43

Dynamics of IP_3 receptor clustering on the endoplasmic reticulum — •Ronny Straube[1,2], Martin Falcke[1], and Michael Ward[3] — [1]Hahn-Meitner-Institut, Glienicker Str. 100, 14109 Berlin, Germany — [2]Max-Planck-Institut für Dynamik komplexer technischer Systeme, Sandtorstr. 1, 39106 Magdeburg, Germany — [3]Department of Mathematics, University of British Columbia, Vancouver, Canada

Motivated by the observation that IP_3 receptor channels (IP_3R) form clusters on the endoplasmic reticulum (ER) during ATP-induced calcium release [1], we calculate the reation rate of small diffusing molecules on a cylindrical membrane by taking into account the cylindrical topology of the tubular ER [2]. The reaction rate is obtained using the method of matched asymptotic expansions. For realistic parameter sets, our calculation predicts clustering rates in the experimentally observed range. Furthermore, it reveals how the cluster rate depends on the relevant system parameters such as the molecule size and the aspect ratio of the membrane. Based on our calculations of the reaction rate, we also study the dynamics of IP_3R clustering as it is triggered by an external calcium signal. A mean-field approach is used to determine the temporal evolution of the cluster-size distribution.

[1] Y. Tateishi et. al., Cluster formation of inositol 1,4,5-triphosphate receptor requires its transition to open state, J. Biol. Chem. 280(8), 6816-6822 (2005).

[2] R. S., Michael J. Ward and Martin Falcke, Reaction rate of small diffusing molecules on a cylindrical membrane, submitted to J. Stat. Phys.

BP 3.12 Mon 17:00 H43

Self-organization of exit sites in the endoplasmic reticulum in mammalian cells — •Matthias Weiss and Stephan Heinzer — Cellular Biophysics Group (B085), German Cancer Research Center, Im Neuenheimer Feld 580, 69120 Heidelberg

Exit sites (ES) are specialized membrane domains of the endoplasmic reticulum (ER) at which cargo proteins of the secretory pathway are packaged into small, COPII-coated vesicles. While the essential COPII proteins that are responsible for the emergence of the vesicles have been identified and characterized during the last decade, their binding kinetics and diffusion properties have remained elusive. Using high-resolution fluorescence microscopy techniques (photobleaching and correlation spectroscopy), we have dtermined the typical exchange time of COPII proteins at single ERES in vivo, the diffusion coefficients of the individual proteins in the cytoplasm as well as the cargo-dependent diffusion of ERES on the ER membrane. We also have quantified the spatial arrangement and size distribution of ERES in vivo. Based on these results, we propose a simple model for the self-organization of ERES that quantitatively matches the experimental data.