BP 57: Membranes and Vesicles I

Time: Thursday 9:30–12:45 Location: H43

Invited Talk BP 57.1 Thu 9:30 H43

Monolayer curvature induced nanoscale structures in lipid membranes — •Friederike Schmid — Institut fuer Physik, Johannes Gutenberg-Universitaet Mainz, Germany

Biological lipid membranes are believed to be laterally heterogeneous and filled with nanoscale ordered "raft" domains. However, the mechanisms stabilizing such small rafts are still under debate. Here we report the observation of raft-like structures of sizes in the order of 10 nm in a coarse-grained molecular model for multicomponent lipid bilayers [1]. Furthermore, we discuss a mechanism that generates nanoscale rafts by a coupling between monolayer curvature and local ordering. The theory rationalizes in a unified manner the observation of a variety of nanoscale structures in lipid membranes: Rippled states in one-component membranes, lipid rafts in multicomponent membranes. Both are observed in our generic simulations, with properties that are compatible with experimental observations [1-3]. Finally, we will discuss the interplay of this mechanism with a similar mechanism based on a coupling between *bilayer* curvature and local ordering, which is well known from the literature and can generate ordered microdomain structures on the scale of 100 nm or more, and show how microdomains can be used to organize nanodomains [4].

[1] S. Meinhardt, R.L.C. Vink, F. Schmid, PNAS 119, 4476 (2013).
[2] O. Lenz, F. Schmid, Phys. Rev. Lett. 98, 058104 (2007).
[3] L. Toppozini et al, Phys. Rev. Lett. 113, 228101 (2014).
[4] L. Brodbek, F. Schmid, AEAM, to appear (2015).

BP 57.2 Thu 10:00 H43

Incorporation of Aescin in DMPC vesicles — •Ramsia Sreij, Carina Dargel, and Thomas Hellweg — Physical and Biophysical Chemistry, Universitätsstraße 25, 33615 Bielefeld

The function of membrane proteins depends on the lipid bilayer thickness (d_z) and its properties. d_z is studied in model membrane systems in form of unilamellar vesicles (ULVs) consisting of 1,2-dimyristoyl-*sn*-glycero-3-phosphocholine (DMPC). DMPC vesicles undergo a phase transition from gel to fluid phase at a temperature $T_M = 23.6°C$. Saponins are a diverse class of natural, plant derived amphiphilic molecules with a peculiar molecular structure made of a hydrophobic scaffold and hydrophilic oligosaccharide chains. They have strong surface activity and are used as natural emulsifiers and foaming agents in food, pharmaceutical and other industries. Their incorporation into the membrane of living cells reduces the cholesterol bioaccessibility by displacement of cholesterol molecules from the bile salt micelles. We studied the saponin Aescin to demonstrate the effect of its incorporation on the lipid bilayer thickness and T_M of small unilamellar DMPC vesicles produced by extrusion. We use small and wide angle X-ray scattering (SWAXS) to measure the effects on d_z in dependence of a varying amount of incorporated Aescin. By differential scanning calorimetry (DSC) and dynamic light scattering (DLS) experiments we show the influence of the saponin incorporation on T_M.

[1] B.A. Brüning, S. Prevost, R. Stehle, R. Steitz, P. Falus, B. Farago, and T. Hellweg, *Biochim. Biophys. Acta*, 1838, 2412-2419.

BP 57.3 Thu 10:15 H43

Squeezing vesicles on a supported lipid bilayer using osmolyte polymer chains: a neutron reflectivity study — •Alexandros Koutsioumpas[1] and Didier Lairez[2] — [1]Jülich Centre for Neutron Science (JCNS), Forschungszentrum Jülich GmbH, Outstation at MLZ, Lichtenbergstrasse 1, 85747 Garching, Germany — [2]Laboratoire Léon Brillouin, CEA/CNRS UMR 12, CEA-Saclay, 91191 Gif-sur-Yvette, France

The approach of two lipid bilayer segments in close proximity is an event of special interest in biophysics, due to its relation with several biological functions such as membrane fusion. Using the neutron reflectometry technique we attempt to gather structural information related to the interaction of a planar supported DPPC bilayer with DPPC vesicles that are "forced" at the interface by the addition of high concentrations of polyethylene glycol chains in bulk solution. At the adopted experimental conditions the major effect that is observed is related to the initial approach of vesicles close to the supported bilayer at temperatures below the liquid/gel transition, followed by their "spreading" and flattening at temperatures above the transition that is attributed to the radical decrease of their bending modulus. The experimental observation is backed up by semi-analytic calculations based on the Helfrich Hamiltonian that describes membrane mechanics. In all cases, no indication of any pre-fusion structure is found. These observations are also discussed in the context of alternative potential approaches for the formation of floating bilayers on a planar surface.

BP 57.4 Thu 10:30 H43

Acto-myosin dynamics drive local membrane component organization in an in vitro active composite layer — •Darius V. Köster[1], Kabir Husain[1], Elda Iljazi[1], Peter Bieling[2], Dyche Mullins[2], Madan Rao[1,3], and Satyajit Mayor[1] — [1]National Centre for Biological Sciences, Bangalore, India — [2]University of California, San Francisco, USA — [3]Raman Research Institute, Bangalore, India

Studies on the organisation of the cell surface have revealed a role for dynamic acto-myosin in membrane protein and lipid organization, suggesting that the cell surface behaves as an active composite. We reconstitute an analogous system in vitro that consists of a fluid lipid bilayer coupled via membrane-associated actin binding proteins to dynamic actin filaments and myosin motors. Varying actin concentration, filament length, and actin/myosin ratio in this minimal system revealed after consumption of a limited ATP pool a phase, characterized by a lattice of polar asters. During the self-organizing aster formation, advection drives transient clustering of membrane components. Increasing levels of ATP produces a constitutively remodelling state of the actin filaments which in turn drive active fluctuations of coupled membrane components, resembling those observed at the cell surface. In a multicomponent membrane bilayer, this remodelling acto-myosin layer contributes to distinct changes in the extent and dynamics of phase segregating domains. These results show how local membrane composition can be driven by active processes arising from acto-myosin which could have implications for the membrane organization in cells.

BP 57.5 Thu 10:45 H43

Structure and dynamics of phospholipid vesicles around the main phase transition — •Beate Brüning[1], Ramsia Sreij[1], Bela Farago[2], and Thomas Hellweg[1] — [1]Bielefeld University, Bielefeld, Germany — [2]Institut Laue-Langevin, Grenoble, France

We use complementary scattering probes to study the molecular and structural reorganization of unilamellar 1,2-dimyristoyl-sn-glycero-3-phosphatidylcholine (DMPC) vesicles undergoing the phospholipids main phase transition. Effects on vesicle morphology and collective shape fluctuations are hierarchically interlinked. We cover length scales from molecule distances to an ensemble of vesicles in solution, and time scales ranging from ns to ms. We find temperature-induced changes in the lipid chain order and bilayer thickness are reflected most strongly in curvature changes and shape fluctuations, rather than in inter-vesicle lipid exchange processes. We comment on the implications for vesicle-membrane budding processes.

[1] Structure and dynamics of phospholipid vesicles around the main phase transition (submitted).

30 min break

BP 57.6 Thu 11:30 H43

Fast tracking of single molecules in live cell membranes at ultra-high resolution with interferometric scattering microscopy (iSCAT) — •Richard Taylor and Vahid Sandoghdar — Max Planck Institute for the Science of Light, Erlangen, Germany

Conventional approaches to track diffusion of lipids and proteins in membranes use fluorescence microscopy, but a low fluorescence rate and inevitable photobleaching severely limit the localisation precision on both the short and long timescales. Furthermore, fluorescence microscopy traditionally also suffers from limited axial resolution. We report on the use of interferometric scattering microscopy (iSCAT) for monitoring of a gold nanoparticle-labelled protein within the live HeLa cell membrane. iSCAT particle tracking has previously demonstrated accurate tracking of labeled single lipids within synthetic membranes to nanometric precision at fast millisecond framerates [1]. Here, we demonstrate use of iSCAT microscopy to track the diffusion of gold labeled transmembrane proteins within the live HeLa cell membrane.

We show that one may track the probe diffusion both in- and out-of-plane, with nanometer-level accuracy and microsecond framerates.

[1] C.-L. Hsieh, S. Spindler, J. Ehrig, V. Sandoghdar, J. Phys. Chem. B. 118, 1545-1554, (2014).

BP 57.7 Thu 11:45 H43

Passive Translocation of Hydrophobic Nanoparticles through a Phospholipid Bilayer — Yachong Guo[1], Emmanuel Terazzi[2], Ralf Seemann[3,4], •Jean-Baptiste Fleury[3], and Vladimir Baulin[1] — [1]Departament d'Enginyeria Quimica, Universitat Rovira i Virgili, 26 Av. dels Paisos Catalans, 43007 Tarragona, Spain — [2]Department of Inorganic and Analytical Chemistry, University of Geneva, 30 quai E. Ansermet, CH-1211 Geneva 4, Switzerland — [3]Universitat des Saarlandes, Experimental Physics, 66123 Saarbruecken, Germany — [4]Max Planck Institute for Dynamics and Self-Organization, Goettingen, Germany

Hydrophobic nanoparticles introduced into living systems may lead to increased toxicity, can activate immune cells or can be used as nanocarriers for drug and gene delivery. The interaction of nanoparticles with bilayers is essential of an in depth understanding of these processes. It is known that small hydrophobic nanoparticles can insert into a lipid bilayer and accumulate in the bilayer core, representing a potential well. Therefore it is generally accepted that escaping the bilayer is unlikely for these nanoparticles. In contrast to this assumption, we demonstrate theoretically how large hydrophobic nanoparticles can cross lipid bilayers with almost no energy barrier, while small hydrophobic nanoparticles stay trapped in the core of the bilayer. This size-dependent translocation was confirmed experimentally using a microfluidic device. Moreover, the kinetic pathway of a single passive translocation event was directly measured and analyzed. (Submitted)

BP 57.8 Thu 12:00 H43

Shaping the Endoplasmic Reticulum network in vitro — •Gernot Guigas, Csilla Ferencz, and Matthias Weiss — Experimental Physics I, University of Bayreuth

Many organelles in eucaryotic cells have complex shapes that deviate significantly from simple spheres. A prime example is the Endoplasmic Reticulum (ER) which forms an extensive network of membrane tubules throughout the cytoplasm of mammalian cells. In order to explore the self-assembly capacity of ER networks we have used an in-vitro reconstitution system and spinning disk confocal microscopy. In particular, we monitored how purified ER microsomes from Xenopus laevis eggs fuse in the presence of purified cytosol, ATP, and GTP. As a result, we observed that a complex network with an ER-like topology and a typical mesh size of some 10 micrometers grew almost instantly on the surface of the incubation chamber. In a subsequent relaxation process, single tubules of the network moved and rearranged until the network had reached a stable configuration. Tubular networks only grew on charged surfaces and they were most stable on surfaces with a medium elasticity. When vesicles were mixed with small silicon oil droplets, tubular networks with a significantly reduced mesh size formed on these droplets, i.e. the ER network structure resembled more closely the native form found in mammalian cells. We conclude from our experiments that ER microscomes have an inherent capacity to self-assemble into a network structure with a mesh size that is influenced by the surface on which the structure grows.

BP 57.9 Thu 12:15 H43

Developing of biomimetic model-membranes to investigate transport processes via X-ray and neutron reflectometry — •Irena Kiesel, Yuri Gerelli, and Giovanna Fragneto — Institut Laue Langevin, Grenoble, France

Transport processes through membranes are fundamental for the biological function of cells in living organisms. To investigate these processes, e.g. transport of drugs into cells, it is necessary to build a reproducible and stable model membrane system, accessible for analytical methods. Typically, model membranes are created as solid-supported lipid bilayers, as they are feasible for surface-sensitive techniques as X-ray- or neutron reflectivity (XRR, NR). In order to allow the penetration of guest molecules through membranes, it is necessary to use a highly hydrated spacer (e.g. polymer brushes). Furthermore, natural membranes are composed by several different saturated and unsaturated lipid species, which is in contrast to normally used model systems, using one or few comercially availible lipid species. Extracted lipids from *Pichia pastoris* yeast are used here in order to model a more natural mimicking membrane. The availability of natural extracts in both hydrogenated and deuterated forms allow the use of the so-called contrast variation method with neutrons. The aim of this project is to create reproducible, stable and tethered model membranes with natural extracted lipids to mimic real membranes and to allow the investigation of transport processes through this membrane. First reults from XRR, NR and other methods (AFM, fluorescence microscopy, QCM) will be presented.

BP 57.10 Thu 12:30 H43

Standing-Wave X-Ray Fluorescence Enables Near-Angstrom Precision Localization of Biologically Important Chemical Elements in Molecular Layers — •Emanuel Schneck[1], Ernesto Scoppola[2,3], Jakub Drnec[4], Cristian Mocuta[5], Roberto Felici[4], Dmitri Novikov[6], Giovanna Fragneto[2], and Jean Daillant[5] — [1]Max Planck Institute of Colloids and Interfaces, Potsdam, Germany — [2]Institut Laue-Langevin, Grenoble, France — [3]Institut de Chimie Séparative de Marcoule, France — [4]European Synchrotron Radiation Facility, Grenoble, France — [5]Synchrotron Soleil, Gif-sur-Yvette, France — [6]Deutsches Elektronen-Synchrotron, Hamburg, Germany

In nature, biomolecules are often organized as functional thin layers in interfacial geometries. The most prominent examples are the biological membranes. But biomolecular layers also play important roles in context with biotechnological surfaces, for instance when they are the result of adsorption processes. For the understanding of many biological or biotechnologically relevant processes, detailed structural insight into the involved biomolecular layers is required. Here, we use standing-wave x-ray fluorescence to determine element-specific density profiles in solid-supported lipid and protein monolayers with near-Angstrom resolution. The technique complements traditional reflectometry experiments which merely yield the layers' "global" density profiles. While earlier work mostly focused on relatively heavy elements, typically ions, we also localize the comparatively light elements S and P, which are found in many biomolecules and therefore particularly interesting.