

membrane hydration dynamics. Contrarily, P181, the most hydrophobic poloxamer known as a membrane permeabilizer, initially penetrates past lipid headgroups and enhances intralayer water diffusivity. Consequently, our results illustrate that the relative hydrophilic/hydrophobic ratio of the polymer dictates its functions.

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Lipid-Polymer Membranes as Carrier for L-Tryptophan: Molecular and Metabolic Properties

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A study of the lipopolymers that encapsulate L-tryptophan was carried out with the main goal of obtaining and characterizing vehicles that could be used as drug delivery systems for the treatment of several metabolic diseases that need an incremented systemic L-tryptophan concentration.

Polymeric liposomes were obtained by UV irradiation of vesicles containing 1,2-bis(10,12-tricosadiynoyl)-sn-glycero-3-phosphocholine (DC8,9PC) and 1,2-dimyristoyl-sn-glycero-3-phosphocholine (DMPC) in 1:1 molar ratio. These polymeric liposomes were also obtained in presence of 10 and 50 mol % of L-tryptophan (respect to total lipid concentration).

Polymerization efficiency in presence of the two mentioned L-tryptophan concentration were studied spectrophotometrically; along with bilayer packing at the polar head region with the probe Merocyanine 540 (MC540). Interaction between lipid-polymer membranes and L-tryptophan was followed by FTIR. Results showed that high L-tryptophan concentration induce the formation of lipopolymers with higher polymeric units, leaded by the higher lipid rigidity adopted in presence of high amino acid concentration. This is a derived implication of the L-tryptophan preferential position interacting at the amine terminal of the choline group.

Stability of lipopolymers with different amounts of L-tryptophan was also studied through release profiles. L-tryptophan release was induced by a concentration gradient and amino acid concentration was determined spectrophotometrically. Polymeric liposomes were able to retain around 80 % of the L-tryptophan after 24 hour. Then, polymeric liposomes with 10 mol % of the amino acid release 5 % more. Nonetheless, retention was high in the elapsed time analysed.

Metabolic activity of the Caco-2 cell line was also studied in the presence of polymeric liposomes with both L-tryptophan concentrations. Cytotoxic effects were low.

In resume, polymeric liposomes studied in this work could be applied as drug delivery systems in order to improve L-tryptophan pharmacodynamics.

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Controlled Cyclic Measurements of Pulmonary Surfactants at Low Surface Tensions

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Pulmonary surfactants cover the alveoli of the lungs and have a vital function in making the process of breathing easy. During inhalation, the surfactant reduces the surface tension of tissue by a factor of about 15. During exhalation, the surface area of the alveoli decreases making the surfactant even more concentrated on the surface. It is known that the highly ordered solid phase of dipalmitoylphosphatidylcholine (DPPC) sustains the near-zero surface tension on the alveoli during exhalation. In order to model the actual surfactant behavior in the alveoli, measurements at near-zero surface tensions are needed.

We have shown before that the compression speed in a Langmuir trough has a distinct effect on the layer formation of DPPC at low surface tensions and temperatures ranging from 20 °C to 37 °C. In this study we further expand this observation by showing controlled cyclic measurements of DPPC at low surface tensions. The measurements were done on ultrapure water surface at temperatures of 20 °C, 30 °C and 37 °C using a Langmuir trough equipped with a ribbon barrier to prevent monolayer leakage. The measurements show reliable compression measurements of natural phospholipid surfactants at surface tensions down to 15 mN/m and demonstrate the usability of the ribbon barrier method. The measurements can be further expanded to examine the phase transitions and the selective enrichment process of DPPC on the alveoli surface.

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Directionality of the Nano-Scale Reversible Collapsed Structures in the Pulmonary Surfactant Film

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Up to now, the determination of the orientation of nano-scaled three-dimensional collapse structures formed at the air-water interface within a compressed Langmuir film was not possible by existing experimental techniques. This is however of special importance if pulmonary surfactant films are investigated, which form reversible surface-associated reservoirs or collapse structures at the air-water interface under dynamic lateral compression and expansion. This surfactant efficient mechanism of collapse has been proposed for a mechanically efficient functioning of the pulmonary surfactant lining, present at the air-alveolar interface. The direction of these reservoirs with respect to the interface, however, has remained dicey since decades. To address this question, we have designed a novel methodological approach to perform bidirectional surface imaging of the pulmonary surfactant harboring nano-scale collapsed structures. This approach has been applied to investigate the collapsed structures formed in a functional analog of the pulmonary surfactant. Here, we prove that the surface-associated structures of the pulmonary surfactant film form towards the air-phase in contrast to the up to now commonly accepted view of an orientation towards the aqueous-phase.

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Membrane Remodelling and Protein Interactions - A Free Energy Perspective

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We investigate the interplay between cell membrane curvature induced at the atomic scale, due to specialized peripheral membrane proteins, and the resulting membrane morphologies, of varying complexity, observed at the meso-scale. The biological membrane, in our approach, is represented by a dynamically triangulated surface while the proteins are modeled as curvature fields on the membrane, that are either isotropic or anisotropic. In order to compare with experiments, we have focused on the ENTH domain containing EP-SIN whose curvature field is modeled as isotropic, and on the BAR, Exo70 and ESCRT family proteins whose curvature fields are determined to be anisotropic, both in experiments and in molecular simulations. Thermal undulations in the membrane and cooperativity in the curvature fields, due to the stabilization of a nematic phase, collectively drive the membrane into different morphological states (buds, tubules, etc.) that resemble those in cellular experiments *in vivo* and vesicle experiments *in vitro*. The relative stabilities of these self-organized shapes are examined by two approaches to compute the free energy of the system: (i) the Widom insertion technique to compute excess chemical potentials and (ii) thermodynamic integration using the Kirkwood coupling parameter to compute free energies. Building on these methods, we propose a hybrid scheme that couples both the approaches for computing free energies in membrane systems with heterogeneous and phase-segregated protein field - examples being the endoplasmic reticulum (ER) membrane discs with α -calreticulin protein confined to the rim and the vesicular bud formed due to the constriction by ESCRT proteins at its neck.

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Quantification of Curvature Gradients in Highly Curved Tubular Lipid Bilayers

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Coupling between membrane shape and composition plays an important role in dynamic life of cellular membranes. It becomes increasingly understood that intrinsic curvature preferences of membrane components, proteins and lipids, provides one of the driving forces for such coupling. However, monitoring curvature-driven sorting of membrane components at physiologically relevant time- and length- scales is a challenging task. Here we propose a new approach for real-time quantification of dynamic gradients of membrane curvature and the associated redistribution of membrane component at nanoscale. This method consists in simultaneous measurements of the electrical conductance and fluorescence of the lumen of lipid nanotubes (NTs). By relating changes in the integral conductance of the NT lumen with those in the axial profile of the fluorescence intensity we obtain the geometrical parameters of the nanotube with 10s of nm precision. Furthermore, by varying the electrical potential applied to the NT membrane we can measure, in real time, changes in the elastic moduli of the NT membrane, e.g. upon adsorption of proteins. For basic lipid compositions, the effective bending rigidity measured here coincides with the published values, while entropy-related correction is evident at high curvature stress.