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The role of phospholipid asymmetry in the transition from the lamellar (L_α) to the in-verted hexagonal (H_{II}) phase upon the temperature increase was considered. The equilibrium configuration of the system was determined by the minimum of the free energy including the contribution of the isotropic and deviatoric bending and the interstitial energy of phos-phospholipid monolayers. The shape and local interactions of a single lipid molecule were taken into account. The minimization with respect to the configuration of the lipid layers was performed by a numerical solution of the system of the Euler-Lagrange differential equations and by the Monte Carlo simulated annealing method. At high enough temperature the lipid molecules attain a shape exhibiting higher intrinsic mean and deviatoric curvatures which fits better into the H_{II} phase than into the L_α phase. Furthermore, the orientational order-ing of lipid molecules in the curvature field expressed as the deviatoric bending provides a considerable negative contribution to the free energy which stabilizes the non-lamellar H_{II} phase. The nucleation configuration for the L_α - H_{II} phase transition is tuned by the isotropic and deviatoric bending energies and the interstitial energy. For the mathematical model the deviations from sphericity of inverted hexagonal phase cross-section were calculated, resulting in lower energy in non-spherical cross-section than in spherical cross-sectin.

1795-Pos Board B639
Phase Behavior And Domain Structures Of Lipid Membranes Under Tension
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The lateral domain structure of lipid membranes is mainly controlled by the thermodynamic characteristics, i.e. composition, temperature and tension. In this study we show experimentally by the combination of fluorescence microscopy and micropipette aspiration techniques and theoretically by phenomenological modeling that the lateral tension of the membrane provides a potent control parameter of the lateral phase behavior and domain structures in lipid membranes. The lateral tension can lead to significant distortions of the phase diagrams and modification of critical behavior, and hence enhancement or suppression of lateral domains.

1796-Pos Board B640
Phosphatidylinositol-4,5-bisphosphate Affects Ceramide 1-phosphate Phase Behavior
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Ceramide is a well-characterized sphingolipid metabolite and second messenger that participates in numerous biological processes. When ceramide is phosphorylated by ceramide kinase (CERK), ceramide-1-phosphate (Cer1P) is obtained. It has recently been proposed that Cer1P is involved in cell survival, cell proliferation, inflammation and phagocytosis. It has been observed that the CERK activity is dependent on the interaction with phosphatidyl inositol-4,5-bisphosphate ($PI(4,5)P_2$). In turn, Cer1P was found to affect the PI3K/AKT pathway. This suggests that $PI(4,5)P_2$ and Cer1P might co-localize and interact. To address this issue, giant unilamellar vesicles (GUVs) composed of POPC and 10% Cer1P were made with different concentrations of brain $PI(4,5)P_2$ (from 2%, 5%, 10% to 20 mol%), labeled with fluorescent lipids and analyzed by fluorescence microscopy. GUVs composed of POPC and 10% Cer1P showed irregular branch-shaped domains, which are characteristic for the Cer1P gel phase. In the presence of 2% brain $PI(4,5)P_2$, the irregularly branched domains took on a shape of beads on strings, i.e., the domains were round indicating increased fluidity. With increasing amount of $PI(4,5)P_2$ added, the bead region became larger and larger. In the presence of 10% $PI(4,5)P_2$; the gel type string regions can barely be seen. For 20% $PI(4,5)P_2$ only one fluid type region can be seen. Since the portion of Cer1P in the GUVs was fixed, (10% of the total lipids) the increasingly larger domains have to be the result of the incorporation of brain $PI(4,5)P_2$ in the Cer1P phase. This incorporation of $PI(4,5)P_2$ in the Cer1P phase leads to an increasing fluidity and increasing size of the domain. In conclusion, Cer1P and $PI(4,5)P_2$ co-localize into a domain when mixed with POPC and this domain exhibits fluid like properties at high $PI(4,5)P_2$ concentrations.

1797-Pos Board B641
Determination of Inter-Phase Line Tension in DMPC/D-Cholesterol mixed Langmuir Films
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The hydrodynamic response of a thin fluid film, whether a Langmuir monolayer at the air/water interface or a cell membrane, is difficult to model, since it involves the coupling of both bulk and surfaces phases. However, such hydrodynamic response is not only intrinsically critical for transport within the layer, it also provides a major available means to evaluate an important parameter for phase-separated layers such as rafts, the line tension. We have developed a line-integral formulation of the hydrodynamic response of phase-separated layers with short-ranged forces, and tested it by comparisons between numerical simulations based on this model and experiment. These experiments both validate the model and demonstrate that the line tension can be determined with unprecedented accuracy and precision. Long-range dipole-dipole interactions are introduced into the model. The method is applied to coexistence between phases in binary phospholipid/cholesterol mixed layers. Data is evaluated for both Brewster and microscopy and fluorescence microscopy and implications of the use of fluorescent probes are discussed.

1798-Pos Board B642
On The Properties Of Surfactant Monolayers At Low Surface Tensions
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The properties of surfactant monolayers at the air/water interface depend strongly on the monolayer surface density. As the density increases, the monolayers undergo transitions from gas to liquid and condensed phases, and transform from 2D to 3D geometry as their stability limit is reached. For a given surfactant, the higher the surface density, the smaller is the monolayer area, and lower is the resulting surface tension at the interface. We found that for selected lipid mixtures and lung surfactant extracts, the monolayer surface tension - area isotherms deviate from the expected dependence. For these mixtures, the captive bubble surfactometer measurements show that at low surface tensions (< 20 mN/m) the reduction of surface tension is accompanied by an increase rather than a decrease of monolayer area. We used a combination of experimental techniques, theoretical models and computer simulations to investigate the properties of monolayers of varying composition at low surface tensions. We hypothesize that the observed effect originates from monolayer 2D-3D transformations. Monolayer wrinkling in particular leads to a decrease of monolayer apparent area and lowers the total surface tension.

1799-Pos Board B643
Effect Of A Water-soluble Polymer On Lamellar Surfactant Phases
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We study the effect of a water-soluble polymer, PEG, on the lamellar phases of different surfactants (non-ionic and ionic). Different experimental techniques (Polarized Light Microscopy, Freeze-Fracture Electron Microscopy, Small-Angle X Ray Scattering, Dynamic Light Scattering, Rheology) show that the polymer strongly affects the structural and physical properties of the membranes. In some cases, the polymer induces a phase of highly packed multilamellar vesicles. We present the effect of polymer concentration and polymer molecular weight.

1800-Pos Board B644
Measuring The Energetic Cost Of Burying An Arginine Sidechain Into A Lipid Bilayer Using A Transmembrane Protein
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Arginine is not a hydrophobic amino acid. On its own, an arginine sidechain would face a large energetic barrier to entry into the apolar core of a lipid bilayer. But, would a similarly high barrier also exist for an arginine if it were part of a whole transmembrane protein? Whether or not that barrier is high, how would such an arginine interact with a lipid bilayer? The answers to these questions will likely have implications on the normal functioning of some ion channels and also on the abnormal mis-folding of some other membrane proteins. Here we attempt to experimentally measure the free energy cost of burying an arginine into the apolar core of a lipid bilayer when that arginine is on the otherwise hydrophobic surface of a transmembrane protein. We also attempt to uncover some molecular details about how the misplaced arginine and its neighboring lipids behave. The transmembrane protein we use is the Outer Membrane Pospholipase A (OmpLa) of *E. Coli*. We engineered OmpLa to have an arginine at each of several positions that, in the crystal structure of

the wild-type protein, face the apolar environment of the lipid bilayer. We then measured the thermodynamic stability of the wild-type protein and of each of the sequence variants by chemical denaturation. We only made these measurements when the proteins appeared to be at reversible equilibrium between folded and denatured states. We also characterized the folded states of each protein by fluorescence spectroscopy and by a functional assay. Those characterizations revealed additional information about how the lipid bilayers may accommodate an arginine.

1801-Pos Board B645

Chain Length Effect on the Association of Fluorescent amphiphiles with lipid bilayer membranes

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The lack of quantitative, kinetic and thermodynamic knowledge regarding the interaction of amphiphiles with lipid bilayers, and the biological and pharmacological relevance of this subject, prompted our group towards a detailed study of those processes [1, 2]. Here we present a detailed study of the interaction of two homologous series of fluorescent amphiphiles (containing one or two acyl chains with different lengths) with a membrane in the liquid disordered phase (POPC). The kinetic rate constants for insertion, desorption, and the corresponding equilibrium partition constants, were obtained. The study was performed as a function of temperature, and the thermodynamic parameters were also obtained.

One of the homologous series studied is a phospholipid labeled with the fluorescent group 7-nitrobenzo-2-oxa-1,3-diazol-4-yl (NBD) in the polar head group and with different lengths of the two acyl chains (NBD-diC_nPE; with n=6, 8, 10, 12 or 14). The other homologous series consists of fatty amines labeled with NBD in the amine group an different *acyl* chain lengths (NBD-C_n; with n=8, 10, 12 or 16).

In contrast to the expectation based on the current model for the transition state in the insertion/desorption process [3, 4], we found a strong dependence between the rate of insertion and the acyl chain length, for both homologous series. The interpretation and implications of the results obtained are discussed.

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1802-Pos Board B646

Effect Of The Acyl Chain Length On The Translocation Rate Of Amphiphilic Molecules In Liquid Disordered And Liquid Ordered Lipid Bilayers

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Passive transport across cell membrane is a significant route for the permeation of xenobiotics through tight epithelia, such as the vascular endothelium that constitutes the Blood Brain Barrier. One of the most important processes for permeation is transmembrane translocation, which depends on the structure of the permeating molecule and on the properties of the lipid bilayer.

In this work we report on the translocation of two homologous series of fluorescent amphiphiles between the two leaflets of lipid bilayers, in the liquid disordered phase (POPC) and in the liquid ordered phase (SpM:Chol 6:4), using established methods [1]. Both series are labeled with the probe 7-nitrobenz-2-oxa-1,3-diazol-4-yl (NBD) in the polar portion and have acyl chains of different length. One of the series is a phospholipid derivative (NBD-diCnPE; n=6,10 or 14) and the other is a fatty amine (NBD-Cn; n=8, 10, 12, 14 or 16). Along these homologous series, the hydrophilic group is maintained and the hydrophilic/hydrophobic ratio is changed *via* the length of the *acyl* chain. The work was done at different temperatures and the thermodynamic parameters were obtained.

For the fatty amine homologous series, the translocation rate constants recovered show a strong dependence on the length of the *acyl* chain for both lipid phases, being very fast for NBD-C8 and almost 3 orders of magnitude slower for the two longer *acyl* chains. A different behavior was found for the phospholipid homologous series, where the translocation was essentially independent on the *acyl* chain length, showing that for this series the solubilization of the polar head group in the center of the bilayer is the higher energetic barrier in the translocation process.

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1803-Pos Board B647

Unraveling the Role of Protein-Proteins Interactions of Annexin at the Membrane

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Protein-membrane interactions are a vital mechanism of propagating signals both across the membrane and between cells. One method of signal propagation is the formation of lipid microdomains that allow the preferential clustering of specific lipid types and proteins. To address this type of signal propagation, we investigated how lipid microdomains form in response to annexin binding to model membranes. Annexins bind to negatively charged (e.g., phosphatidylserine [PS]) membranes in a calcium-dependent manner and lead to the formation of PS-enriched microdomains in supported planar bilayers. Two distinct mechanisms of signal propagation via protein-lipid binding are addressed. First, we hypothesize that proteins can transmit binding information via the ordering of the lipid acyl chains upon binding. Alternatively, we predict that when a protein binds a specific lipid preferentially, protein-protein interactions are enhanced on a membrane surface. The role of lipid acyl chain ordering and protein-protein interactions as distinct mechanisms of signal propagation through lipid binding will be illustrated.

1804-Pos Board B648

Artificial Phospholipid Bilayers On Nano-patterned Gold Surfaces For Biosensing

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As cell surface mimics, supported lipid bilayers are suitable as functional overlayers that enable the study of binding interactions that occur at cell surfaces. These interactions are relevant to cell-cell interactions, and pharmacological applications. Their use however, is limited by the types of surfaces they can reliably be assembled on. We demonstrate the assembly of artificial phospholipid bilayers on gold substrates patterned with a regular array of nano-holes. The lipid layers are characterized by imaging and force indentation using an atomic force microscope. We also demonstrate a biosensor that combines nano-hole arrays, and lipid bilayers.

1805-Pos Board B649

Fission Of Lipid Nanotube By Osmotic Pressure

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Formation of new membrane compartments, such as transport vesicles in cells, is finalized by scission of the membrane connection between the vesicle and the parent membrane. To avoid leakage of the vesicle contents, fission has to pass through so called hemifission state, where inner monolayer of membrane neck self-merge while outer preserves its continuity. Creation of hemifission is coupled to generation of high membrane curvature by specialized protein machinery. To reveal the intrinsic behavior of lipid bilayer in this process we studied protein-free fission of membrane nanotubes (NT) subjected to osmotic stress. As expected, lowering of the osmolarity of the external solution caused NT expansion while increasing of the osmolarity produced NT narrowing. We found that osmotic pressure could squeeze NT to a critical radius where non-leaky fission occurred spontaneously. Furthermore, when we progressively increased the amount of cholesterol in the NT membrane to augment its rigidity, the value of the critical radius remained unchanged (corresponding to the lumenal radius of approximately 2 nm). Thus we conclude that membrane rearrangements leading to non-leaky membrane fission can be initiated by a critical narrowing of the membrane tubule.

1806-Pos Board B650

Measurement Of Mechanical Parameters Of Lipid Bilayer Form The Deformation Of Membrane Nanotube In Electric Field

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The effect of a longitudinal electric field on the shape of a lipid nanotube formed in an electrolyte solution is considered experimentally and theoretically. Application of a moderate (50-250mV) potential difference between two ends of the nanotube caused the tube expansion so that its shape deflected from the initial cylindrical to the parabolic one. The magnitude of this deviation depends on 1) the potential difference applied, 2) initial lateral tension and 3) bending modulus of the nanotube membrane. This deviation can be quantified as an effective radius of the nanotube determined by the mechanical parameters of the nanotube membrane and the magnitude of the applied electrical field. From the dependence of this radius on the potential difference the values of