deprotonation of PI(4,5)P_2 due to association of the cations. This effect was significantly larger in the presence of Ca^{2+}. Multilamellar vesicles containing PC and PI(4,5)P_2 with varying amounts of cholesterol were also studied. The 4- and 5-phosphates of PI(4,5)P_2 were found to have a significant downfield shift in the presence of 40 mol% cholesterol. The cumulative effects of cholesterol in combination with the common inner leaflet phospholipids, PE and PI, were also examined.

1468-Pos Board B198
Lipids as Regulators of Effective Membrane Rigidity
Ksenia Chekashkina[1], Peter Kuzmin[1], Pavel Bashkirov[1], Vadim Frolov[2,3].
[1]A.N. Frumkin Institute of Physical Chemistry and Electrochemistry, Moscow, Russian Federation, [2]Biophysics Unit (CSIC, UPV/EHU) and Department of Biochemistry and Molecular Biology, University of the Basque Country, Leoia, Spain, [3]IKERBASQUE, Basque Foundation for Science, Bilbao, Spain.
The rigidity of lipid bilayer, the structural core of all cellular membranes, is one of the key force factor in membrane remodeling. For most of ''biomimetic'' lipid compositions the rigidity is thought to be only a weak function of the composition. However, for multicomponent membranes the composition is coupled to geometry, resulting in lateral redistribution of components in curvature gradients. Such redistribution can substantially facilitate local membrane deformations. We detected that such a decrease of apparent membrane rigidity in pure lipid bilayers containing physiological amounts of dioleoylphosphotidylethanolamin (DOPE), the lipid characterized by highly negative intrinsic curvature. Analyzing fast shape transformations of membrane nanotubes containing different amounts of DOPE we found that the membrane softening followed concentration-dependent redistribution of DOPE towards negative membrane curvature. The apparent bending rigidity of DOPE-containing membranes decreased almost twofold at 30mol% of DOPE, indicating that similar amount of DOPE in cellular membranes can substantially facilitate deformations.

1469-Pos Board B199
Enhancement in Lipid Bilayer Partitioning of Lysolipids and Fatty Acids Induced by their Composition
Radha Ranganathan, Jasmeet Singh.
California State University Northridge, Northridge, CA, USA.
Distribution of the solutes: lysopalmitoylphosphatidylcholine (LPPC), Palmitic acid (PA) and their 1:1 mixtures between water and dipalmitoylphosphatidylcholine (DPPC) bilayer were determined using a fluorescence probe that selectively detects these solutes in water. Membrane phase as well as solute concentration itself affects partitioning. Water solute concentrations were obtained at each of several bilayer lipid concentrations between 10 and 400 μM, from slopes of the linear variation of probe fluorescence properties with total solute concentrations of up to 10 % of the solvent lipid concentration. Dynamic Light Scattering experiments confirmed that the lipid/solute aggregates were vesicles in this range. Lipid concentration dependence of the solute component in water was fit to a thermodynamic model of solute distribution between two coexisting solvents. Water/bilayer partition coefficient and the solute transfer free energy were determined from the fit. Main findings are: (1) Water to bilayer transfer free energy of solute is lower for 0 to 2 % solute mole fraction than for 2 to 10 %, signaling composition induced bilayer relaxation that increases bilayer solubility, beginning at 2 % solute mole fraction. (2) Partition coefficients are in the order LPPC>PA> $LPPC^+PA$ at 37 °C. The enhanced partition coefficient of $LPPC^+PA$ signifies synergism toward increased solubility in the bilayer-gel phase. Enhancement effects were not present, where the DPPC bilayer is in the liquid phase, The observed order in the partition coefficients, at 50 °C, was LPPC≈$LPPC^+PA$>PA. The behavior of the partition coefficients in the gel and liquid phase is similar in character to the observed presence of synergism in the transmembrane permeabilityin the bilayer gel phase and lack of it in the liquid phase. The present results provide experimental evidence that increased presence of solutes in the membrane also enhance transmembrane permeability.

1470-Pos Board B200
A Comprehensive Study of Preferential Interaction of Cholesterol and its Fluorescent Analogs with Different Classes of Phospholipids
Shishir Jaikishan, Thomas K.M. Nyholm.
Biochemistry, Department of Biosciences, Åbo Akademi University, Turku, Finland.
Cell membranes are composed of various glycerophospholipids and sphingolipids. Cholesterol is believed to interact with these phospholipids to modulate the fluidity of the membrane. We have introduced a novel approach to measure the preferential interaction of cholesterol with different classes of phospholipids having different head-groups and acyl-chain compositions. Phospholipids involved in the study are phosphatidylcholine (PC), phosphatidylglycerol (PG), phosphatidylethanolamine (PE), phosphatidylserine (PS) and sphingomyelin (SM). These phospholipid classes constitute the bulk of the membrane phospholipids. The acyl chain configurations of the saturated and unsaturated glycerophospholipids involved in this study are di 16:0 and 16:0(*sn1*)-18:1(*sn2*) respectively. Correspondingly, the *N*-linked acyl chain configurations in SMs are 16:0 and 18:1(Δ9-cis). Affinity of sterol analogs cholestatrienol and bodipy-cholesterol towards phospholipid bilayers is also studied to compare their affinities with cholesterol. Large unilamellar vesicle (LUV) systems with (sterol) donor vesicles and (sterol) acceptor vesicles are used. Acceptor LUVs are allowed to accept the sterol from donor vesicles, transfer being assisted by methyl-β-cyclodextrin. Diphenylhexatriene-phosphatidylcholine (DPHPC) was used as a fluorescent probe in cholesterol donor acceptor anisotropy experiments. We observed that cholesterol showed different affinity for phospholipids as compared to its fluorescent analogs. Affinity of cholesterol and the fluorescent analogs for different phospholipid bilayers depended not only on the head groups of the phospholipids but also on their acyl chain configuration.

1471-Pos Board B201
Vesicles and Phase Dynamics: Cross-Linking Effects
Michael S. Kessler, Susan Gillmor.
Chemistry, The George Washington University, Washington, DC, USA.
We study lipid phase behavior using giant unilamellar vesicles to model cell membrane dynamics. Vesicles allow us to isolate the lipid rearrangement due to cross-linking, common activity on cell surfaces. Biotylated lipids, avidin and its analogues allow us to model lipid rearrangement due to cross-linking at the headgroup position, where cross-linking is the linking of two molecules (biotinylated lipids) via a cross- linking agent (avidin). Using phase specific dyes, we study the changes that occur with the addition of a cross-linker to the system. Förster Resonance Energy Transfer (FRET) enables us to detect phase changes on the submicron scale, beyond the limits of conventional microscopy. Using FRET we detect lipid rearrangement associated with the transition from one-phase vesicles to two-phase vesicles using two different fluorescent dyes, a donor and acceptor. From this simple cross-linking system, we model membrane responses to protein complex formation and oligomerization.

1472-Pos Board B202
Domain Size Distribution in Phase Separated Cholesterol/Phospholipid Langmuir Monolayers: Line Tension and Transition Kinetics
Emil Eldo[1], Andris Bibelnieks[2], Promise Okeke[3], Joan C. Kunz[1], **Benjamin L. Stottrup**[2].
[1]Chemistry, Augsburg College, Minneapolis, MN, USA, [2]Physics, Augsburg College, Minneapolis, MN, USA, [3]Biology, Augsburg College, Minneapolis, MN, USA.
Multicomponent phase separated phospholipid monolayer systems of a canonical cholesterol and DMPC (~30/70 mole percent) have been used to study several aspects of cholesterol/phospholipid interaction and phase behavior. Despite the successful characterization and theoretical approaches applied to these systems there are still important details of monolayer morphology that are not fully understood. Here, we address the role of transition kinetics on domain size distributions. For our experiments, three different barrier speeds were chosen (4, 40, 400 cm squared per minute) as the monolayer passed through the miscibility phase transition (8.4 mN/m transition pressure). Average domain size was observed to decrease as the barrier speed increased (transition rate). The detailed size distribution measurements also provide the opportunity to measure changes in phase fraction and size distribution with monolayer surface pressures (2, 4, and 6, mN/m). Careful study of factors influencing the size distribution of phase separated domains is particularly relevant to the recently proposed line tension measurement technique (Lee et al., vol. 108 pp. 9425, PNAS 2011). We have implemented this method for the monolayer system studied here. A comparison to previously implemented line tension measurements based on a Fourier analysis of boundary fluctuations approaches will be presented. Finally, a brief comparison on the role of dyes and dye quality will be presented. Comparison among one year old Texas Red and new Texas Red, showed an impact in the sizes of domains.