The presence of less than 20% D-PUFA in the total amount of PUFA was shown to completely stop LPO in yeast cells under oxidative stress. Here, we report results of studying the impact of phospholipids containing D-PUFAs on LPO in model bilayer lipid membranes by using two approaches: 1) monitoring diene conjugates in liposomal membranes and 2) measuring permeabilization of liposomes. The latter was estimated with the fluorescence correlation spectroscopy method from changes in the amplitude of the autocorrelation function of liposomes loaded with the fluorescent dye sulforhodamine B. Both approaches revealed a strong protective action of small amounts of deuterated PUFA against LPO in bilayer membranes. The extent of protection depended on the nature of D-PUFAs, being determined predominantly by the total level of deuterated bis-allylic (CD2) groups. For each D-PUFA, there was found a threshold percentage (about 20-25% for 1-acyl-2-(11,11-D_2-linoleyl)-*sn*-glycero-3-phosphatidylcholine and much less for 1-acyl-2-(6,6,9,9,12,12,15,15,18,18-D_{10}-docosahexaenoyl)-*sn*-glycero-3-phosphatidylcholine) in the H-PUFA matrix that inhibited LPO, which could be attributed to an ability of D-PUFA-derived radicals to interrupt LPO chain reactions in a lipid bilayer. The protecting effect of D-PUFAs was compared to the action of various free-radical scavengers, e.g. trolox. This work was supported by the Russian Science Foundation grant 19-74-00015.

1130-Pos
Permeability of Human Red Blood Cell Membranes to Hydrogen Peroxide
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Hydrogen peroxide (H2O2) and other reactive species are important physiological mediators in the vascular system. Enzymatic production of H2O2 is involved in regulating cell growth, proliferation and vasodilation. Whereas endothelial cells are important sources of H2O2, red blood cells (RBC) are considered the most important sinks of H2O2 in the vasculature. However, little is known about the permeability of their membrane to H2O2. The permeability coefficient of human RBC membranes to H2O2 was determined using the enzyme latency method, based on measuring the rate of H2O2 decomposition in lysed vs whole cells. If the passage through the membrane is the rate limiting step in H2O2 decomposition, then a difference is observed that can be used to calculate the permeability coefficient. Additional experiments were done to differentiate between simple diffusion through the lipid fraction and facilitated diffusion through protein channels. The lack of reported permeability coefficients for lipid membranes prompted us to do experiments with phospholipid-cholesterol liposome membranes that indicated that simple diffusion is a slow process. Determination of partition coefficients in different solvents mimicking different depths of the membrane indicate that the low permeability of lipid membranes to H2O2 is caused mainly by its very low solubility in the acyl region of the bilayer. The activation energy of permeation through RBC membranes suggested that protein channels were involved in facilitating H2O2 diffusion through the membrane. Inhibitors of hAQP3 and hAQP1 had no effect in H2O2 consumption rate, suggesting that other membrane proteins may be involved. Although the RBC membrane presents a significant barrier to H2O2 passage, especially in comparison with other solutes such as oxygen and nitric oxide, the permeability is still high enough to support the role of RBC as sinks of H2O2 in circulation.

1131-Pos
Counterintuitive Electrostatic Forces in Liposome Colloidal Crystals
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We previously studied the repulsive electrostatic forces among like-charged liposomes in deionized water by measuring the shear moduli (rigidity, stiffness) of liposome-water suspensions as a function of liposome charge and concentration (Cohen, Wei & Ou-Yang). The liposomes were extruded, unilamellar, 100 nm, made of weak-acid POPG and zwitterionic POPC mixtures in incremental ratios. At low charge the shear moduli increased with liposome PG content as expected, reaching a maximum at ~20% PG, then decreased with further increase of PG. At 100% PG the suspension was flaccid with a shear modulus near zero. Our measurements show that in deionized water weakly-charged liposomes interact strongly, and strongly-charged liposomes interact weakly. Here we analyze this counterintuitive finding. We use a Debye-Hückel (DH) Wigner-Seitz (WS) model to calculate electrostatic potentials and electric fields in the suspensions analytically. The unusual bulk behavior arises from a strict correlation, mandated by global electroneutrality, between the deprotonated PG liposome charge and the number of solvated H^+ ions in a WS cell. We map the conditions for weak vs. strong inter-liposome forces in terms of the H^+ screening length relative to the WS sphere radius, which is a measure of the mean distance between liposomes. The H^+ screening length depends on the solvated H^+ mean concentration, hence on the net liposome charge including charge-regulation effects. The liposome surface potentials are shown to be small, which justifies use of the DH approximation. We plot electrostatic-potential and pH profiles in a WS cell as a function of the liposome PG/PC ratio, calculate PG/PC at the crossover from strong to weak coupling, and determine the added-salt concentration required to mask the above effects. These results may be relevant to protein-protein interactions in low ionic-strength electrolytes.

1132-Pos
Effect of Styrene Maleic Acid Copolymer Length on Biological Membrane Solubilisation and Properties of Native Nanodiscs
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The styrene-maleic acid copolymer (SMA) has proven to be a valuable tool in the study of membrane proteins because it can be used to isolate membrane proteins in native nanodiscs. Of the commercially available SMA copolymers, the one with the smallest average weight (Xiran 30010) was shown to be the most efficient for biological membrane solubilisation. However, Xiran 30010 still has a relatively large average size and a broad size distribution. Less polydisperse and smaller SMA fragments have been isolated and tested on phospholipid vesicles(Domínguez Pardo et al., 2018) but not on biological membranes. Here we prepared such smaller SMA fragments to assess the amount of solubilisation of *E. coli* membranes with over-expressed KcsA by turbidity assays and SDS-PAGE analysis. The size of the native nanodiscs was analyzed by SEC, EM and dynamic light scattering and the stability of proteins in the nanodiscs was studied using spectroscopic methods. The results show that there is an optimal size for membrane solubilisation, which is different from the situation in model systems where the smallest polymers are better solubilisers. The results provide new insights into the mode of action of SMA copolymers and will be helpful for optimising conditions to solubilise and characterise membrane proteins in their native environment.

1133-Pos
Demixing in Membranes and their Encapsulated Solutions
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Liquid-liquid phase separation drives a range of biophysical processes in many locations throughout cells, from the interior of nuclei to outer membranes. It is known that phase separation in a cell's interior creates 3D liquid droplets composed of proteins, which are sensitive to the system's physiochemical conditions. In membranes, such as those of giant plasma membrane vesicles (GPMVs) and living vacuole membranes, phase separation is two dimensional. The composition of lipids and cholesterol in the membrane determines the temperature (T_{mix}) at which the system transitions from a mixed to phase separated system. When 3D droplets and membranes physically touch, actin-independent endocytosis can occur. Together, these observations lead to the question: How does demixing in the solution affect demixing in the membrane? We investigate this question using giant unilamellar vesicles (GUVs) and an aqueous two-phase system of polyethylene glycol and dextran.

Posters: Membrane Dynamics II

1134-Pos
Observations of Compound Penetration in *Escherichia coli* using Ethidium Bromide as a Model Compound
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According to the Centers for Disease Control and Prevention, approximately 2 million people are afflicted with antibiotic-resistant infections annually. One of the contributing factors to a bacterium's resistance to antimicrobials is the presence of multidrug efflux pumps. By removing various toxins from the host cell (i.e. dyes to antibiotics), these pumps ensure the survival of the pathogen. Little is known regarding the kinetics of these pumps. Found in virtually all types of bacteria, there are multiple families of efflux pumps. This research focuses on AcrAB-TolC, an active efflux pump that is commonly found in Gram-negative bacterium such as *Escherichia coli* (*E. coli*). The overall objective of this research is the development of a mathematical model to accurately represent compound penetration into *E. coli* and the contribution of efflux in the process. Using multiple concentrations of the fluorescent compound ethidium bromide (EtBr) and stopped-flow spectrometry, observations of changes in fluorescence intensity over time were made using