

dependence of the transition temperature on the DOPC/POPC ratio. Notably, a continuous shift to higher temperatures with increasing DOPC content was observed for macroscopic domains. Our data suggest that the domain size influences all structural parameters as well as the melting behavior.

1101-Pos Board B169

Role of Binding Free Energy in Membrane Bending by Protein Crowding Gokul Raghunath, Brian Dyer.

Chemistry, Emory University, Atlanta, GA, USA.

Steric confinement of asymmetrically distributed proteins has been shown to drive membrane bending with or without a physiologically native binding interface. While various models have been proposed to explain this phenomenon, a thorough physical basis for how the energetics of protein binding couples to the changes in the membrane morphology still remains unclear. A number of questions pertaining to the mechanism still persist. What is the role of membrane's physical properties in regulating protein density? How does receptor density transiently change the affinity of protein binding to the lipid surface? What are the key factors that pay for the entropic cost of bending membranes? To address these questions, we utilize a simple model system comprised of Histidine-tagged green fluorescent protein and nitrilotriacetic acid modified lipid molecules, to provide evidence suggesting that the free energy of protein binding directly couples to membrane deformation. Investigating the concentration dependence of the protein binding to the membrane surface, we observe interesting changes in the protein binding modality as the protein density increases. We postulate that changes in protein density on the membrane surface can affect the overall membrane tension and fluctuation, which, in turn can help the proteins overcome the large energy barrier to bending the membranes. These results advance our understanding of the mechanism underlying membrane bending induced by protein crowding.

1102-Pos Board B170

Anionic H-Bonds in the Chlorosulfolipid Surface Bilayer of *O. danica* the Strength of a Bacterial Cell Wall

Robert E. Parrish¹, Thomas H. Haines^{2,3}, Robert G. Hohenstein⁴.
¹Chemistry, Stanford University, Palo Alto, CA, USA, ²Biochemistry, Rockefeller University, New York, NY, USA, ³Chemistry, CCNY of CUNY, New York, NY, USA, ⁴Chemistry, CCNY of CUNY, New York, NY, USA.

Ochromonas danica, a protozoan algal di-flagellate grows in fresh water bogs at pH 4.3. Despite the absence of a wall, its bilayer membrane protects it from osmotic bursting. Lacking P-lipids, its flagellar membrane is made of single-chain (C_{20}) poly-chlorinated 1,14-disulfates with up to 6 chloros. The C_{14} -sulfate on each chain forms an H-bonded dense anionic surface sheet attracting H_3O^+ (hydronium ions) to the membrane fresh-water surface. Each H_3O^+ ion can enter the low dielectric and H-bond to the chloros spaced along the chains, due to chloro-electronegativity. The field created by the anionic hydrophobic- C_{14} -sulfate sheet deep in the bilayer attracts the H_3O^+ ions toward it. The sulfate-H-sulfate H-bonds, or anionic H-bonds (AHBs) are much stronger than those same bonds at the C_{14} -sulfate sheet, because the C_{14} -sheet faces water, which weakens AHBs. Both sulfate sheets require AHBs to strengthen the bilayer but the C_{14} -sulfate sheet is significantly stronger because of its aprotic environment. (Sigala, et al. JACS (2015)137,5730). Like the single-chain bilayers of the chlorosulfolipids, single-chain oleic acid bilayers use a COOH AHb and also have a second binding domain in the low dielectric, a $\Delta 9$ double bond. We use it to compare energy measurements. *O. danica*'s surface bilayer also contains unique PUFAs, dominated by arachidonic, linoleic, and linolenic acids, which further strengthen the bilayer.

1103-Pos Board B171

Ring and Tails: Exploring the Intimacy of Cyclodextrin - Membrane Interactions

Monika Kluzek, Fabrice Thalmann, Marc Schmutz, Carlos Marques. Institut Charles Sadron, CNRS, Strasbourg, France.

Cyclodextrins (CDs) are cyclic, ring-shaped molecules made of sugar moieties, consisting of a hydrophobic cavity and a hydrophilic outer part. Such a structure allows CDs to form inclusion complexes with hydrophobic molecules, where the guest molecule is physically entrapped in the host's (CD) cavity. This feature finds a wide range of applications in drug delivery, pharmaceutical and food industries, as inclusion complexes with CDs show enhanced bioavailability and longer circulation times.

Recently, α CDs have been reported to directly interact with aliphatic tails of phospholipids from biological and model bilayer membranes. As the tails are not directly accessible for molecules from the aqueous solution, this observation suggests complex interplays and possible membrane remodeling occurring at the CD/membrane/water interface. Interestingly, despite the wide applications of CDs, the essentials of this process and its possible consequences in vivo remain poorly understood.

Herein, we use a combination of complementary biophysical techniques to uncover the inner workings of interactions between CDs and model lipid membranes, focusing on CDs' membrane-perturbing properties. Specifically, we employ Quartz Crystal Microbalance (QCMB-D) to reveal variations in viscoelastic properties of a lipid membrane induced by the presence of α CD. Furthermore, the application of confocal microscopy and CryoTEM allows us to explore the role of membrane curvature-mediated interactions between α CDs and the lipid membrane.

1104-Pos Board B172

Influence of Cholesterol on Lateral Segregation in Bilayers Containing Different Sphingomyelins and Unsaturated Phospholipids

Oskar Engberg, Victor Hautala, Thomas K.M. Nyholm, J. Peter Slotte. Faculty of Science and Engineering, Åbo Akademi University, Turku, Finland.

Unsaturated and saturated phospholipids (PLs) may laterally segregate into ordered and disordered domains. Cholesterol is known to influence this lateral domain formation in model membranes, which likely resembles the formation of cholesterol rich nanodomains in biological membranes. Cholesterol prefers interacting with saturated PLs over monounsaturated and especially polyunsaturated PLs. Cholesterol also favors interacting with sphingomyelin (SM) over saturated phosphatidylcholines. We have earlier shown that the relative cholesterol affinity for unsaturated and saturated PL can determine to what degree cholesterol promotes lateral domain formation. The fluorescence decays of *trans*-parinaric acid (tPA) was analyzed to detect how much SM was required to form ordered domains in a fluid bilayer. We determined lateral segregation in bilayers containing 0 or 20 mol% cholesterol to compare to what degree cholesterol promoted lateral segregation. Monounsaturated and polyunsaturated PLs were chosen as fluid lipids. As ordered lipids SMs with different *N*-acyl chain lengths and saturation were chosen. These SMs were compared to SM mixtures, both specifically chosen mixtures and biological mixtures. Sterol partitioning experiments were performed to determine the relative cholesterol affinity for the saturated and unsaturated PLs. Differential scanning calorimetry was used to determine if cholesterol could stabilize domain thermostability in complex bilayers. The preliminary results showed that cholesterol promoted lateral segregation for all SMs studied, including the mixtures. General observations about how cholesterol influence domain formation will be presented. These, can be of importance for understanding dynamics of nanodomains in biological membranes.

1105-Pos Board B173

The Molecular Structure of Sphingomyelin in Fluid Phase Bilayers Determined by the Joint Analysis of Neutron and X-Ray Scattering Data

Frederick A. Heberle¹, Milka Doktorova², Jianjun Pan³, Drew Marquardt¹, Richard W. Pastor⁴, Richard M. Venable⁴, Norbert Kucerka⁵, John Katsaras¹.

¹Biology and Soft Matter Division, Oak Ridge National Laboratory, Oak Ridge, TN, USA, ²Weill Cornell Medical College, New York, NY, USA, ³University of South Florida, Tampa, FL, USA, ⁴National Institutes of Health, Rockville, MD, USA, ⁵Joint Institute for Nuclear Research, Frank Laboratory of Neutron Physics, Dubna, Russian Federation.

We have determined the fluid bilayer structure of palmitoyl sphingomyelin (PSM) and stearoyl sphingomyelin (SSM) by simultaneously analyzing small-angle neutron and X-ray scattering data. We report structural parameters obtained using the newly developed scattering density profile (SDP) model for sphingomyelin lipids, including the area per lipid, total bilayer thickness and hydrocarbon thickness, in addition to lipid volumes determined by densitometry. Unconstrained all-atom simulations of PSM bilayers at 55°C using the C36 CHARMM force field produced a lipid area of 56 Å², a value that was 10% lower than the one determined experimentally (61.9 Å²). Furthermore, scattering form factors calculated from the unconstrained simulations were in poor agreement with experimental form factors, despite the fact that segmental order parameter (S_{CD}) profiles calculated from the simulations were in relatively good agreement with previously published S_{CD} profiles obtained from NMR experiments. Conversely, constrained area simulations at 61.9 Å² resulted in good agreement between the simulation and experimental scattering form factors, but not with S_{CD} profiles. We discuss possible reasons for the discrepancies between these two important types of data that are frequently used as validation metrics for molecular dynamics force fields.

1106-Pos Board B174

Investigation of Transbilayer Coupling in Gel-Fluid Asymmetric Lipid Vesicles

Barbara Eicher^{1,2}, Drew Marquardt^{1,2}, John Katsaras³, Georg Pabst^{1,2}.

¹Institute of Molecular Biosciences/Biophysics division, University of Graz, Graz, Austria, ²BioTechMed, Graz, Austria, ³Biology and Soft Matter Division, Oak Ridge National Laboratory, Oak Ridge, TN, USA.

Biological membranes display an asymmetric lipid distribution along the bilayer normal. Membrane asymmetry is also thought to affect the bilayer