

the effect of glycerol on the surfactant - lipid system in terms of surfactant self-association (critical micelle concentration, CMC), membrane partitioning (partition coefficient K and α_{Hmc}), and the onset of membrane solubilization (i.e., bilayer-to-micelle transition, characterized by a specific surfactant-to-lipid mole ratio in the membrane denoted $Resat$) by isothermal titration calorimetry (ITC). One effect expected for glycerol is its tendency to 'salt out' hydrophobic molecules. This promotes all aggregation phenomena (K increases, CMC decreases) but has little effect on the balance between the aggregates ($Resat \sim const.$). Furthermore, glycerol gradually dehydrates the polar head group, which renders the effective molecular shape more favorable for a bilayer (K increases) whereas micellization is much less affected ($CMC \sim const$) so that bilayers are stabilized compared to micelles ($Resat$ increases). Our results indicate that the behavior of the sugar based surfactant octyl glucoside seems governed by the salting out effect, whereas for ethylene oxide surfactant C12EO8, headgroup dehydration seems to be the key effect explaining the effects of glycerol.

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Synthetic and Mycobacterial Trehalose Glycolipids Confer Dehydration Resistance to Supported Phospholipid Monolayers

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We have recently demonstrated that the glycolipid trehalose dimycolate (TDM), a major outer membrane component of dehydration resistant *Mycobacterium tuberculosis* (MTb), can impart significant dehydration resistance to supported phospholipid membranes. We now report studies of other, related glycolipids both natural and synthetic that exhibit behavior similar to TDM, conferring protection from desiccation to membranes of which they are constituents. We examined solid-supported lipid monolayers, characterizing membrane integrity and two-dimensional fluidity before and after de- and re-hydration with fluorescence imaging and fluorescence recovery after photobleaching (FRAP). As with TDM, the degree of protection is dependent on the fraction of synthetic lipid in the monolayer and there is a distinct minimum fraction needed for protection by each glycolipid. We can control the synthetic lipid fluidity and minimum protecting fraction by designing and synthesizing lipids with particular hydrophobic chain lengths, saturation, and branching, thereby illuminating the role of molecular structure in biophysical function. The advent of these synthetic, protective glycolipids opens the door to the creation of lipid bilayers and liposomes since the relevant hydrophobic and hydrophilic domain sizes can be controlled. These new structures allow investigations of the physical origins of well-known mycobacterial properties beyond dehydration resistance in controlled experimental contexts, such as the inhibition of membrane fusion.

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How Small Polar Molecules Protect Membrane Systems Against Osmotic Stress

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We investigate how small polar molecules, urea and glycerol, can act to protect a phospholipid bilayer system against osmotic stress. The osmotic stress can be caused by a dry environment, freezing, or through exposure to aqueous systems with high osmotic pressure due to solutes like in saline water. A large number of organisms regularly experience osmotic stress and it is a common response to produce small polar molecules intracellularly. We have selected two ternary systems of urea-water-dimyristoylphosphatidylcholine (DMPC) and glycerol-water-DMPC as model systems to investigate the molecular mechanism behind this protective effect, and we put a special emphasis on applications in skin care products. Using solid-state NMR, DSC, X-ray diffraction and sorption-microbalance measurements we study the phase behavior of the lipid system both exposed to an excess of solvent of varying composition and for systems exposed to water at reduced relative humidities. In this we have arrived at a rather detailed thermodynamic characterization. The basic findings are: i) In excess solvent the thermally induced lipid phase transitions are only marginally dependent on the addition of urea(glycerol). ii) For lipid systems with limited access to solvent the phase behavior is basically determined by the amount of solvent irrespective of the urea(glycerol) content. iii) The presence of urea (glycerol) have the effect to retain the lipid in liquid crystalline phase down to low relative humidities (64% for urea, 75% for glycerol at 27°C), whereas the transition to the gel phase occurs already at a relative humidity of 94% in pure water, demonstrating the protective effect of the polar molecules against osmotic stress. iv)

In skin care products urea and glycerol are referred to as a moisturizer, which we find slightly misleading as it replaces the water while keeping the physical properties unaltered.

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How Bilayer Curvatures Modulate Molecular Reaction Efficiencies In A Membrane Junction

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Chemical reactions between ligand/receptors taking place in the intermembrane space between opposing lipid bilayers are crucial for maintaining processes vital for eukaryotes and multicellular organisms, e.g. vesicle trafficking and immune responses. The mechanism of chemical reactions taking place in bulk are understood in terms of molecular collective properties such as pressure, temperature and concentration, whereas a reaction taking place between bilayers proceed by more subtle pathways. The properties of the membranes into which the receptors are embedded modulate reaction efficiencies and should be understood in terms of contact area, receptor density and interbilayer forces.

We here present results from a single-vesicle based binding assay showing how the curvature of two lipid bilayers drastically alters binding efficiencies. We studied three biochemical binding reactions (i) trans-SNARE complexation, (ii) streptavidin/biotin recognition and (iii) calcium-mediated lipid chelation all taking place in the confined space of a membrane junction. We measured binding probabilities as a function of membrane curvatures and found that the probability of successfully completing the binding reaction could be changed by up to two orders of magnitude depending on the bilayer shape.

We built a simple model based on receptor membrane density, electrostatic bilayer repulsion and membrane shape to account for the observation. We believe the presented assay and model to have an impact on the understanding of biological recognition reactions taking place across a membrane junction, such as vesicle trafficking and fusion, as well as neuronal and immunological synapse formation.

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Effect of Poloxamer 188 on the Osmotic Response of Cell Membranes

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Poloxamer 188 (P188), an amphiphilic triblock copolymer of poly(ethylene oxide)-block-poly(propylene oxide)-block-poly(ethylene oxide) has been shown to effectively interact with injured plasma membrane and restore its function both *in vitro* and *in vivo*. Elucidation of the mesoscopic and molecular mechanism that mediates the interaction between this triblock copolymer and damaged membranes will help to improve the current approach in development of Poloxamers for therapeutic purposes. Previous work done by our group examined the interaction between P188 with phospholipid monolayers and demonstrated that P188 inserts selectively into damaged portions of the membrane and corrals surrounding lipids. Upon restoration of membrane integrity, the inserted polymer is squeezed out. Here, the effect of P188 on membrane permeability under osmotic stress was investigated by using giant unilamellar phospholipid vesicles, the simplest biomimic membrane which retains the essential curved bilayer structure. Results will be presented detailing how the osmotic gradients and P188 concentration affect P188 in corralling membrane lipids.

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Self-assembly Formation of Multiple Tethered Lipid Bilayers

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Tethered lipid bilayers have been proven powerful as experimental models in studies of membrane-spanning proteins, which are currently the most important targets in drug discovery. Tethering of planar lipid membranes reduces the influence from the solid support on the lateral mobility of the membrane constituents and provides a sufficiently large solvent reservoir underneath the membrane for studying molecular transport events. Inspired by cell-cell junctions, where membrane residing proteins control the separation between two or more membranes without interfering with their integrity, we developed a new self-assembly route for formation of multiple macroscopically homogeneous and highly fluid tethered lipid bilayers (lipid diffusivity $\sim 5 \mu m^2/s$) with compartmentalized inter-membrane volumes geometrically confined by membrane-anchored DNA duplexes. The formation of multiple macroscopically homogeneous planar membrane-membrane junctions with sealed inter-membrane liquid reservoirs was accomplished using so called bicelles, which is a versatile class of model membranes generally composed of a mixture of the long-chained dimyristoyl phosphatidylcholine (DMPC) and the short-chained dihexanoyl PC. Quartz crystal microbalance with dissipation (QCM-D) was