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The Therapeutic Role of Recombinant Human MG53 Protein in Wound Healing
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MG53 is a member of tripartite motif (TRIM) family of proteins and an important component of the membrane repair machinery. In skeletal muscle, MG53 protects against eccentric contraction related damage, while in cardiac muscle MG53 protects against ischemia-reperfusion injury. While previous studies establish MG53 function in striated muscles, little is known about MG53 function in other tissues. Here we investigate the function of MG53 in the skin and its contribution to wound healing. We found that loss-of-function of MG53 caused delayed wound healing and defect in hair follicular structure with the mg53-/- mice. In vitro studies with expression of GFP-MG53 in cultured keratinocytes showed that the protein responded to membrane damage in the same fashion as in muscle fibers and other non-muscle cells. In vivo studies using rodent models of incisional and excisional wound demonstrated that both topical and subcutaneous application of recombinant human MG53 (rhMG53) protein could improve wound healing. For improvement of the efficacy of rhMG53 in wound healing, we used a formulation where rhMG53 is packaged into a hydrogel in liquid format at room temperature, which can polymerize upon exposure to the body temperature for controlled delivery of rhMG53 to the wound site. This hydrogel formulation significantly improved the efficacy for rhMG53 in healing of incisional wound. Our data suggest that MG53 has strong potential as a therapeutic agent for wound healing.

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The Unique Roles of Hybrid Lipids in Lipid Membrane Domain Size and Order
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Hybrid lipids (HL) are phospholipids with one saturated and one unsaturated chains. Although it is believed that hybrid lipids can act as linactants (i.e., 2D surfactants) in cell membranes to reduce line tension of lipid domains, as a group, their fundamental roles in membrane heterogeneity have not yet been systematically investigated. In this study, three hybrid lipids of different degrees of chain unsaturation (i.e., 16:0-18:1PC (POPC), 16:0-18:2PC (PLPC), and 16:0-20:4PC (PAPC)) are compared in their abilities to alter the composition, order and compactness of lipid domains. The liquid-ordered and liquid-disordered (Lo+Ld) phase boundaries of HL/di18:0PC(DSPC)/cholesterol systems were determined from giant unilamellar vesicles (GUV). We found that the Lo and Ld lipid domains in PAPC/DSPC/CHOL and PLPC/DSPC/CHOL mixtures are micron-sized, and only POPC/DSPC/CHOL system has nanoscopic domains. The results show that some poly-unsaturated HLs essentially behave like the double-unsaturated di18:1PC (DOPC), and the mono-unsaturated POPC clearly displays both properties of unsaturated lipid and linactant. The obtained phase boundaries indicate that both POPC and PLPC have significant partition in the Lo phases. Our MD simulations reveal that these hybrid lipids decrease the order and compactness of Lo domains. Thus, hybrid lipids distinguish themselves from other lipids in this combined ''partitioning and loosening'' ability in Lo domains, and this ability could facilitate membrane proteins to partition into lipid-raft-like domains in biomembranes. Our line tension measurement and Monte Carlo simulation both show that even in the best cases, HLs are weak linactants with only modest ability in reducing line tension. Furthermore, the reduction is through two separated mechanisms: one through reducing the compositional differences of Lo and Ld domains, and the other through weak linactant action.

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Cardiolipin Localisation in Buckled Membranes
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The spatial organisation of lipids and proteins in a cell membrane is often of critical importance for function of cell processes and to generate and maintain the cell shape. Some proteins, for example, localise to high curvature regions, while some do so showing a dependence on the local concentration of a particular membrane lipid component. Curvature sensing and generation by lipids and proteins is a potential driving force for the segregation of the cell membrane components. It has been experimentally shown that cardiolipin (CL) localises to the cell poles of E. Coli. The shape of CL indicates a preference for negative curvature which could contribute to localisation. In order to gain a deeper insight into membrane curvature sensing we develop a computational method to study this phenomenon. The basic idea is to simulate buckled membranes with a range of curvatures and collect statistics of CL positions. We perform coarse-grained molecular dynamics simulations of a lipid bilayer consisting of the phospholipids PG, PE and CL, with proportions corresponding to E. Coli. We indeed find that CL localises to regions of negative curvature in a way that can be described by a simple phenomenological theory of membrane curvature sensing.

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Towards Far-Field Microscopic Imaging of Supported Lipid Bilayer Optical Anisotropy
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The characterization of lipid bilayers is of great importance to biology as they are omnipresent in cellular organisms. Frequently, fluorescent dyes are used to observe and characterize lipid membranes.[1] However, often questions can be raised regarding potential unintended interference of fluorescent dyes with the biological system under investigation. Polarimetry and in particular imaging ellipsometry has proven to be a non-disruptive characterization technique for supported lipid bilayers.[2]
Here, a novel label-free characterization scheme for supported lipid bilayers (SLB) is presented, based on the accurate determination of spatially resolved optical anisotropy. Experimental feasibility is demonstrated for spatially resolved optical anisotropy measurements of supported lipids bilayers over a large ~200μm^2 field of view with microscopic resolution. Through a combination of spectroscopic imaging ellipsometry and liquid phase atomic force microscopy, non-destructive optical characterization of lipid bilayers without the use of external markers such as fluorescent labels is achieved. Optical anisotropy results for a DOPC/DOPS 4:1 lipid mixture are in-line with other experimental observations, but are limited in experimental error due to the presence of excess noise. Possible hard- and software improvements to achieve experimental noise reduction are contemplated.
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The Effect of Membrane-To-Domain Thickness Mismatch in Phase Separation Ternary Lipid Systems as a Function of Vesicle Size
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Cellular membranes are no longer viewed as a homogeneous mix of lipids and proteins, but rather as having distinct lipid domains, so-called ''rafts'', which are key for many biological processes. Much work devoted to understanding the actual mechanisms that drives lateral organization in cell membranes has been done on model membrane systems. This approach has given great insight into the formation of lipid rafts because in simple ternary mixtures with a saturated lipid, an unsaturated lipid and cholesterol, a region of liquid-liquid coexistence was found, with one of the liquid phases rich in cholesterol and saturated lipids, the finger-print of rafts. However, there is still no clear understanding of the molecular parameters that drives phase separation. Recently a new curvature hypothesis proposed a correlation between composition, leaflet coupling and emulsion-like induced curvature to predict phase separation and the formation of domains. To explore these ideas, we recently studied the phase behavior of well-researched phase separating ternary mixtures: dDPPC-DOPC-Cholesterol (1:1:1) and dDMPC-DOPC-Cholesterol (1:1:1), in 30nm vesicles using Small Angle Neutron Scattering. dDMPC is a 14-carbon long lipid while dDPPC is 16-carbon long lipid. As temperature was lowered, the system with dDPPC showed excess scattering, whereas the system with dDMPC showed no scattering even below its melting temperature. These results are interesting when compared to the previously established work where both systems show phase separation in GUVs as seen by fluorescent microscopy. Phase separation behavior differences as a result of variations to the tail length of the saturated lipids in our vesicles with high curvature gives insight into the role of local curvature (emulsion-like effects) at the domain-membrane interface.