Biomolecular Interactions at the Lipid Interface

by

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Abstract

Lipids constitute the major component of cell membranes, forming bilayers which act as an ion-impermeant barrier. Nature has exploited the self assembling tendencies of lipids, along with the diverse array of different lipid types with unique properties including charge, reactivity, ligand presentation, curvature, and packing density within the bilayer to generate surfaces with highly controllable spatially resolved properties. This resolution is further enhanced by the ability of lipids within a structure to separate into individual phases, capable of forming multiple isolated regions with specific properties even within a single bilayer. The existence of these rafts is controversial, however much evidence exists demonstrating that cells utilize these lipid properties to cluster receptors, isolate specific regions of the membrane, and present ligands in a multivalent manner. Given the diversity and importance of lipid bilayers in nature, it is desirable to better understand the properties of these bilayers, especially in the context of how they interface with other biomolecules. A better understanding of these protein-lipid interactions can be used to develop better nanomaterials, design new drug delivery systems, and understand the pathology of numerous diseases characterized by cell membrane leakage. To this end, this work initially describes the use of a phase separated liposome with specific enzyme adsorption regions which is capable of stabilizing enzymes under denaturing conditions by preventing lateral interactions on the soft support surface. This work demonstrates a nature inspired method for designing surfaces for interfacing proteins with nanomaterials while enhancing their resistance to nonnative conditions. The next section of this document describes the ability of specific amyloid aggregates to increase membrane permeability. Using planar bilayer electrophysiology, the duration of the toxic species of different serum amyloid a (SAA) variants was studied, demonstrating that different SAA variants formed bilayer permeabilizing species with drastically different persistence times. Furthermore, the mechanism by which different forms of SAA permeabilize the bilayer was shown to be different for two isoforms with a high degree of sequence homology. The Alzheimer's related beta amyloid peptide was also studied in order

to determine a potential specific oligomer responsible for bilayer permeabilization which may be responsible for cell death in vivo. The presence of an oligomer specific antibody was also shown to inhibit permeabilization in a dose dependent manner. Finally, we describe the proteolytic activity of serum amyloid A 2.2, and investigate it's effect on fibronectin, an extracellular matrix protein. Using electrophoresis and mass spectrometry, these experiments suggest that SAA may be able to directly degrade fibronectin, a major extracellular matrix protein associated with cell adhesion. This work demonstrates that knowledge of lipid-protein interactions both at the bilayer surface and with the bilayer interior is important to a number of fields such as nanomaterials, biocatalysis, drug delivery, and obtaining a better understanding of amyloid diseases.