New and Notable

Breaking Up Isn't So Hard to Do

Anne K. Kenworthy
Departments of Molecular Physiology and Biophysics & Cell and Developmental Biology, Vanderbilt School of Medicine, Nashville, Tennessee

Lipid rafts are among the best-studied, yet least understood features of cell membranes. Most researchers agree that lipid rafts are a type of membrane domain that contains mixtures of cholesterol and either phospholipids or sphingolipids with high melting temperatures (1). Such lipid mixtures exhibit both high order and rapid lateral motion of their components and are therefore referred to as liquid-ordered phases (L_o) to contrast them with lipids in liquid-crystalline/liquid-disordered (L_d) states. The hypothesis that cell membranes contain a mixture of L_o and L_d domains has spawned intense study of the phase behavior of lipids that support L_o/L_d phase separation in artificial bilayers (2). One of the most striking features of the lipid domains observed in artificial membranes is their ability to grow to spectacularly large sizes. However, study after study has failed to detect large-scale domains in living cells. Even when scrutinized using highly sophisticated imaging approaches, most cholesterol-dependent domains appear to be, at most, on the order of 5–10 nanometers in size under steady-state conditions.

What makes cell membranes so recalcitrant to forming micron-scale lipid domains? On page 3113 of this issue, Yethiraj and Weisshaar use an in silico approach to test the hypothesis that the presence of obstacles precludes the formation of large lipid domains (3). They postulate that integral membrane proteins attached to the cytoskeleton act as immobile obstacles. Using an Ising model of a binary lipid mixture whose phase behavior is tuned to match that of a canonical lipid raft-forming mixture (1:1:1 palmitoyl-sphingomyelin, dioleoyl phosphatidylcholine, and cholesterol), they show that the effect of the inclusion of obstacles is to depress the temperature at which phase separation occurs. The model predicts that for this lipid mixture, fluid-fluid phase separation normally occurs at 37°C. However, in the presence of obstacles, domains are broken up into nanoscale clusters, reducing the tendency of the lipids to phase separate. Remarkably, as little as 10% coverage of the bilayer by obstacles is sufficient to cause such behavior.

The findings of Yethiraj and Weisshaar (3) thus support a role for cytoskeletally bound proteins as negative regulators of lipid domain formation. This model predicts that in the absence of the cytoskeleton, domain formation should be enhanced, in excellent agreement with recent experimental studies showing that L_o-like/L_d-like domain separation can occur in plasma membrane blebs devoid of the cytoskeleton (4). The model further raises the interesting possibility that the mean spacing of obstacles in cell membranes can influence the size of lipid domains.

In addition to demonstrating a role of proteins in determining the properties of L_o/L_d domains, the results of this study provide a tantalizing clue as to how the distribution of proteins with respect to domains may be controlled. To date, most experimental studies have focused on partitioning into the liquid-ordered versus liquid-disordered phase. In such assays, transmembrane proteins and peptides tend to show a strong preference for partitioning in L_d domains, whereas GPI-anchored proteins prefer an L_o-like phase. However, the existence of a class of proteins that act as ''two-dimensional surfactants,'' helping to stabilize the interface of L_o and L_d domains, has also been postulated by a number of workers in the field. Consistent with this possibility, Yethiraj and Weisshaar (3) find that in their simulations, obstacles tend to fall at interdomain boundaries, and speculate that this is a function of their energetic neutrality in the model. It will be of interest to determine how the attractive and repulsive interactions between proteins and lipids further shape the size and shape of lipid domains using the types of modeling approaches described here in future work.

Is the presence of obstacles the only reason lipid rafts are not readily observed in vivo? A number of alternative models remain to be tested. For example, cell membrane lipid composition may not support fluid-fluid phase immiscibility (2). In addition, triggering events may be required to coalesce and stabilize small rafts. Indeed, this notion has become so widespread that it was incorporated into a recently proposed formal definition of rafts: ''Small rafts can sometimes be stabilized to form larger platforms through protein-protein and protein-lipid interactions'' (1). The size of lipid domains in cells is also likely to be controlled by features of cell membranes not easily mimicked in artificial membranes such as membrane turnover, the high concentrations of membrane proteins in cells, and the complex composition of cellular membranes (2,4). There is also building evidence that some types of microdomains, such as nanoclusters enriched in the small GTPase Ras or GPI-anchored proteins, are actively regulated to maintain a constant fraction of clustered to monomeric molecules over a wide range of expression levels. Clearly, with the development of new modeling and experimental systems to probe membrane domain structure, lipid rafts cannot remain an elusive entity for much longer.

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Address reprint requests to A. K. Kenworthy, E-mail: anne.kenworthy@vanderbilt.edu.

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