Research Highlight #147

Lipid domains in Intact Fiber-Cell Plasma Membranes: structure, dynamics, and function

Witold K. Subczynski[a], Marta Pasenkiewicz-Gierula[a,b], Marija Raguz[a,c], Laxman Mainali[a], James S. Hyde[a]
[a]National Biomedical EPR Center, Dept. of Biophysics, [b]Medical College of Wisconsin; Dept. of Biophysics & Biotechnology, Jagiellonian University, Krakow, Poland; [c]Dept. of Medical Physics & Biophysics, University of Split, Croatia

Introduction: Age-related changes in lipid composition of the human fiber-cell membrane of the eye lens are much greater than age-related changes in membranes of other organs and tissues. The phospholipid (PL) composition changes with increase of sphingomyelin and depletion of phosphatidylcholine. The cholesterol (Chol) saturation level of PLs also increases from a Chol:PL molar ratio of 1:2 in the cortex to a ratio as high as 4:1 in the lens nucleus. At this high content, Chol saturates the bulk PL bilayer and induces formation of cholesterol domains. Additionally, aged fiber-cell membranes become loaded with proteins. The organization of membrane proteins changes with age, although these changes do not cause loss of lens transparency.

Methods: For details, see our recent review [1]. We tested the hypothesis that high Chol content and the presence of cholesterol bilayer domains (CBDs) are necessary to maintain lens-membrane homeostasis throughout life. Both spin-labeled phospholipids and spin labeled cholesterol were used in the study.

Results: Chol mobility in membranes saturated with Chol is similar to that of the surrounding PL bilayer and is vastly different in the rigid structure of Chol crystallites. Formation of CBDs precedes formation of Chol crystals [2] as indicated in Fig.1. CBDs are supported by the surrounding PL membrane saturated with Chol. A threshold is observed at about 66 mol%. Below this threshold, a structured single-phase region exists (l_o with CBD). Above this threshold, a two-phase region is found where the "l_o with CBD" phase coexists with Chol crystals. Fig. 2 illustrates the presence of the four purported lipid domains in intact fiber-cell membranes of the human eye lens. We confirmed the existence of bulk, boundary, and trapped lipid domains [3]. However, we have not been able to discriminate the CBD, which was easily detected in cortical and nuclear lens lipid membranes [4]. We quantitatively evaluated the relative amounts of PLs and Chol in lipid domains in intact human eye-lens membranes. Our molecular dynamics (MD) simulations confirmed EPR results [5].

Implications: It seems likely that CBDs are precursors to formation of Chol crystals. At 50 mol%, the size of the individual domains increases with increase of Chol content. Above the threshold, the CBD cannot be further supported by the surrounding PL bilayer and the CBD collapses from the two-dimensional structure of the domain to the three-dimensional structure of the monohydrate Chol crystal. Lipid domains formed in the presence of membrane proteins, namely boundary and trapped lipid domains, account, respectively, for 37 and 46% of total PLs in cortical and nuclear membranes. EPR and saturation recovery methods indicate virtually total exclusion of Chol from lipid domains formed in the presence of membrane proteins in cortical intact membranes. However, in nuclear membranes, about 50% of Chol is located in the protein-induced lipid domains. The amount of lipids in domains uniquely formed due to the presence of membrane proteins is greater in nuclear membranes than in cortical membranes.

Discussion: Human lenses can be different not only because of age, but also because of varying health history of the donor. Recently we extended our methods for the first time to samples from a single donor, which enables the study of changes in fiber-cell membranes occurring with age and cataract formation.

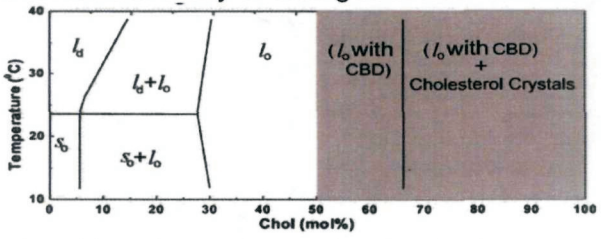

Fig. 1. Phase diagram for cholesterol/DMPC mixtures.

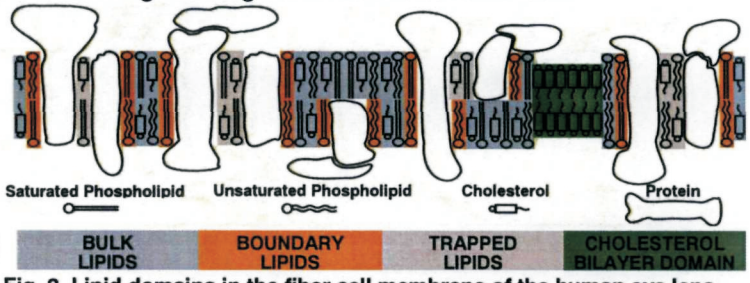

Fig. 2. Lipid domains in the fiber cell membrane of the human eye lens.

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