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Theodore L Hazlett and Enrico Gratton.

Punctuated lipid mass loss during enzyme-mediated lipid hydrolysis on giant unilamellar vesicles.

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Abstract

Giant unilamellar vesicles (GUV) have been successfully used as model membranes in a variety of lipid-lipid and lipid-protein systems, including the binding and subsequent hydrolysis of GUV membrane lipids by secreted phospholipases A2 (sPLA2). Hydrolysis of GUV lipids by sPLA2 commonly results in the steady decrease in GUV size with the eventual disappearance of the GUV altogether. However, we have found that lipid loss is not always gradual but can be distinctly punctuated with sudden GUV diameter changes as great as 14%. Clearly, this effect is the result of the competition between membrane forces that tend to stabilize the bilayer structure, and the destabilizing forces introduced by the buildup of hydrolysis products, lysolipid and free fatty acid. Concomitant with the observed lipid loss is an abrupt increase in Laurdan GP and, in one case, the apparent expulsion of an internal lipid vesicle. A GP increase is indicative of greater lipid order. Interestingly, this GP effect is observed simultaneously across the entire GUV, which suggests a global membrane impact. Data are discussed in terms of lipid lateral pressures, product buildup and the possible formation of lipid structures on the GUV membrane. Support: NIH, PHS P41 5 RR03155.