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Authors

Sanchez, S
Gratton, E
Bagatolli, L
et al.

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Susana A Sánchez, Enrico Gratton, Luis A Bagatolli, and Theodore L Hazlett.

Phospholipase A2 association and hydrolysis of homogeneous- and mixed-lipid giant unilamellar vesicles.

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[Abstract](#), [Internal PDF](#)

The secreted phospholipase A2s (sPLA2) are interfacial enzymes which act at the surface of lipid bilayers. Membrane curvature, phase, charge, and packing have all been claimed to have direct influence on sPLA2-membrane interactions. To address issues pertaining to sPLA2 - membrane interactions, we have used giant unilamellar vesicles (GUVs) as model membrane systems. The GUVs have been doped with membrane probes in order to track PLA2-dependent vesicle morphology and fluidity changes. In addition, a fluorescently tagged sPLA2 (from *Crotalus atrox* venom) has been made and utilized to directly observe sPLA2 binding to the membrane surface. In the presence of active enzyme, vesicles of POPC and DPPC (liquid crystalline phase) displayed a steady shrinking, indicating lipid loss from the bilayer presumably due to absorption of hydrolysis products. Interestingly, DMPC vesicles displayed sPLA2-dependent morphology changes, a size decrease, and the formation of smaller vesicles inside the original GUV. Our binding data show that Ca^{++} , or the Ca^{++} mimic Ba^{++} , is required for membrane binding (POPC GUVs, 20°C). Binding and activity of sPLA2 with mixed phase vesicles, such as DPPC:DPPE (7:3, at 53°C), suggest that PLA2 associates primarily with the DPPC-predominate domains. Financial support: NIH RR03155, American Heart Association-IL 97-CGS-07/GIO.