

Incorporation of Triacylglycerol and Cholesteryl Ester Droplets in Phase-Separated Giant Unilamellar Vesicles

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SUPPORTING INFORMATION

Formation of Vesicles Consisting of a Single-Bilayer Region and a Double-Bilayer Region. We sometimes observed a vesicle consisting of a double-bilayer spherical segment and a single-bilayer spherical segment (Figure S1). We previously investigated such membrane structures obtained after the incorporation of triacylglycerol droplets in phosphatidylcholine vesicles in a single fluid phase labeled with Texas Red 1,2-dihexadecanoyl-*sn*-glycero-3-phosphoethanolamine, triethylammonium salt (TR-DHPE).¹ In our previous study, the two membrane regions could be readily distinguished by their different TR-DHPE fluorescence intensities. However, in the present study, it was sometimes difficult to identify two-region vesicles. The vesicle bilayers consisted of liquid-ordered and liquid-disordered phases, which were labeled with different concentrations of TR-DHPE and 1,2-dipalmitoyl-*sn*-glycero-3-phosphoethanolamine-*N*-(7-nitro-2-1,3-benzoxadiazol-4-yl) (ammonium salt) (NBD-DPPE) (Figure S1, magenta and green). Thus, it was not possible to determine the lamellarity based solely on the difference in the TR (or NBD) fluorescence intensities. Because of this difficulty, their membrane structures were not analyzed.

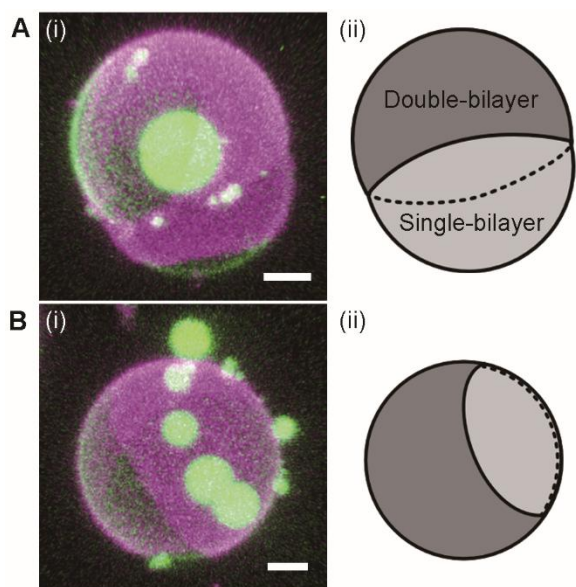


FIGURE S1. (A) and (B) Vesicles consisting of double-bilayer and single-bilayer regions obtained after the incubation of DOPC/DPPC/cholesterol (3:3:2) GUVs labeled with 0.1% TR-DHPE (magenta) and 0.2% NBD-DPPE (green) with triicosenoin droplets labeled with 0.005% TopFluor-TG (green). (i) Confocal fluorescence images and (ii) the corresponding membrane structures. Scale bars, 5 μm .

Reference

- (1) Kataoka-Hamai, C. Triacylglycerol-Droplet-Induced Bilayer Spontaneous Curvature in Giant Unilamellar Vesicles. *Biophys. J.* **2024**, *123* (13), 1857–1868