Buffer-Dependent Self-Assembly and Multiscale Structure of POPC Vesicles Revealed by SAXS and WAXS

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Lamellarity and structure of lipid vesicles arise from an interplay between interfacial forces and preparation conditions. Here, the influence of buffer system (Tris, sodium phosphate, HEPES), concentration (10–100 mM), and extrusion pore size (30 and 50 nm) on POPC vesicle self-assembly is investigated.
Small-angle X-ray scattering (SAXS) reveals pronounced differences in lamellarity. Vesicles in sodium phosphate buffer show coexistence of uni- and multilamellar populations, consistent with the competition between hydration repulsion and attractive interbilayer forces controlling bilayer stacking [1,2]. In contrast, low-concentration Tris yields predominantly unilamellar vesicles, whereas HEPES promotes multilamellar structures, suggesting specific buffer–membrane interactions. Membrane thickness decreases from ~38 Å to ~34 Å with increasing concentration, consistent with electrostatic screening and changes in headgroup orientation [1–3].
Dynamic light scattering (DLS) shows that vesicle size depends strongly on buffer conditions for 50 nm extrusion, while this dependence is reduced for 30 nm pores, highlighting the interplay between chemical environment and geometric confinement. This agrees with established extrusion methods [4,5] and recent findings that incomplete bilayer separation can preserve multilamellar structures [6]. Cryo-TEM confirms the SAXS results, showing mixed lamellarity in sodium phosphate buffer and predominantly unilamellar vesicles in low-concentration Tris. Wide-angle X-ray scattering (WAXS) probes lipid packing and headgroup organization, linking molecular structure to vesicle morphology.
Overall, buffer composition and extrusion conditions determine vesicle self-assembly across length scales and provide a framework for preparing biomimetic membranes with a high fraction of unilamellar vesicles.

Keywords:
lipid vesicles, self-assembly, lamellarity, SAXS, bilayer structure, buffer effects.

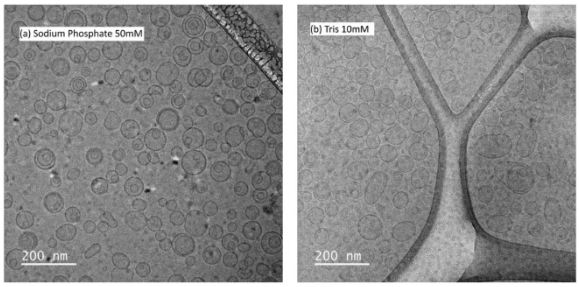

Figure 1: Cryo-TEM micrographs of POPC vesicles extruded through 50 nm membranes. (a) Sodium phosphate buffer (50 mM) showing coexistence of unilamellar and multilamellar vesicles. (b) Tris buffer (10 mM) showing predominantly unilamellar vesicles.

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