Not-so-model systems: Quantification of multilamellar sub-populations in phospholipid vesicles with increasing cholesterol

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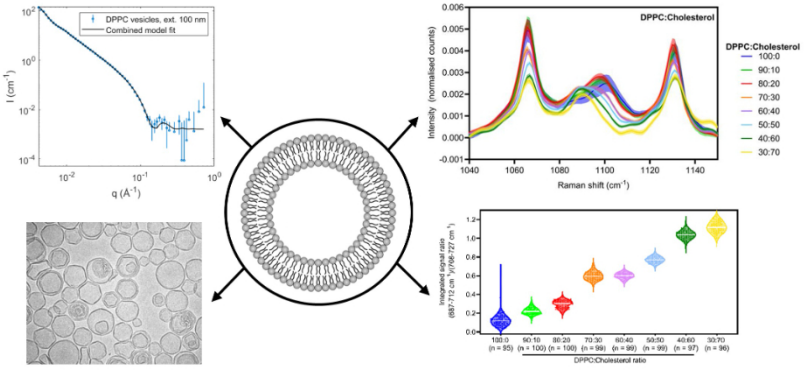
Vesicles, particles composed of one or more lipid bilayer(s) enclosing an aqueous core, have found applications in a wide variety of research areas. This includes models of biological membranes, as drug delivery vectors, and to reconstitute and study membrane proteins, among many others (Pick, et al., 2018). The vesicle lipid composition can vary from simple one-component systems to complex multi-component biomembranes with leaflet asymmetry, with cholesterol highlighted as a key component (Doktorova, et al., 2025). Vesicles have a simple structure and well-established preparation protocols, therefore formulations are often assumed to contain a monodisperse distribution of uniformly spherical unilamellar particles. It has previously been shown, however, that multilamellarity strongly depends on the lipid composition and preparation method (Nele, et al., 2019).
In this project, we have studied the structure of vesicles prepared via extrusion and composed of i) DOPC, ii) POPC, or iii) DPPC with varying amounts of cholesterol on the single particle level. Using single particle Raman trapping analysis (SPARTA), the PC/cholesterol composition variation between individual particles, the saturation limit of cholesterol, and changes in lipid ordering, such as the transition from gel phase to liquid disordered phase for DPPC, were clearly visible. The shape, size, and structure of the vesicles were characterised by small-angle neutron scattering (SANS) and cryogenic transmission electron microscopy (cryoTEM). Comparison of the SANS and cryoTEM data enabled us to develop a combined model containing unilamellar, bilamellar, multilamellar, and elongated particles, which was required to fully describe the data.
In this work, we demonstrate the prevalence of multilamellar vesicle sub-populations even in the simplest one- and two-component systems. This can have important consequences for healthcare applications including vesicle-mediated delivery of nucleic acid therapeutics.

Keywords:
small angle neutron scattering, vesicle, drug delivery, cryoTEM, single particle analysis



Summary of the vesicle characterisation approaches including SANS, cryoTEM and SPARTA for the example of DPPC.

References:

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