mRNA-lipid nanoparticles as a vaccine platform: structural effect of cholesterol substitution with an adjuvant lipid

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Lipid nanoparticles (LNPs) are the leading delivery system for mRNA vaccines. LNPs are composed of a cationic ionizable lipid, a phospholipid, cholesterol, and a pegylated lipid. A major challenge remains the dose-dependent reactogenicity, primarily due to the ionizable component. Despite the clear potential of mRNA vaccines, the structure-function relationship of mRNA-LNPs remains relatively unexplored. Only a few studies have investigated the structure and lipid organisation in mRNA-LNPs and correlated it to their biological function; particularly with the clinically approved ONPATTRO® formulation based on DLin-MC3-DMA, which displayed a core-shell sphere structure. [1,2]
Here, we substitute cholesterol with monomycoloyl glycerol (MMG-1), which is a known adjuvant and stabiliser, potentially enabling reduction of the dose of ionisable lipid and hence reactogenicity. Promising results from our collaborators show that MMG-1 stabilises mRNA-LNPs without reducing vaccine potency. [3]
We investigated mRNA-LNP formulations based on the cationic ionisable lipidoid C12-200, the phospholipid DOPE, pegylated lipid DMPE-PEG, and cholesterol, and a range of formulations where cholesterol was replaced by MMG-1 to reveal the effect on particle shape, structure and lipid arrangement.
We employed small-angle x-ray and neutron scattering (SAXS and SANS) in a range of temperature and pH values to mimic relevant physiological conditions. We have used deuterated cholesterol, deuterated C12-200, and deuterated MMG-1 to investigate the component distribution in mRNA-LNPs.
mRNA-LNPs displayed shape and internal structure slightly dependent on the lipid composition but highly affected by the presence of mRNA cargo and by the pH conditions. Overall, this study contributes to the understanding of the composition-structure relationship in mRNA-LNPs. We aim to use this knowledge for the rational design of safer and more effective mRNA vaccines.

Keywords:
mRNA-lipid nanoparticles, small-angle scattering

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