LIPID NANOPARTICLES FOR EGFP-mRNA DELIVERY IN VITRO

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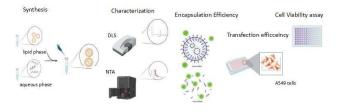
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Abstract

To express functional proteins in target cells, messenger RNA (mRNA) therapeutics require efficient, biocompatible delivery systems. Lipid nanoparticles (LNPs) have emerged as better alternatives to polymeric and viral systems. This study evaluates LNPs formulated with a relatively new ionizable lipid, 4A3-SC8, and the benchmark lipid D-Lin-MC3-DMA, for the in vitro delivery of EGFP-mRNA in adherent cells. Both formulations were characterized using dynamic light scattering and nanoparticle tracking analysis to determine size, polydispersity and concentration. Encapsulation efficiency was quantified using the RiboGreen® assay; delivery efficiency evaluated by EGFP-fluorescence in A549 cells; and cell viability was assessed via the MTT assay. Compared with D-Lin-MC3-DMA LNPs, 4A3-SC8 LNPs enhanced transfection efficiency and mRNA expression with no significant difference in cell viability across five concentrations. These findings underscore the impact of structure and lipid composition on functional delivery efficiency, and support 4A3-SC8 as a promising ionizable lipid for mRNA therapeutics.

Keywords: Lipid nanoparticles, ionizable lipids, SC8 lipid, MC3 lipid, mRNA delivery, in vitro, delivery efficiency, encapsulation efficiency, viability

Graphical Abstract

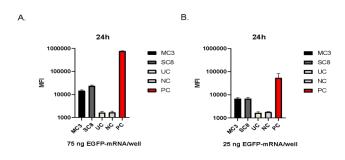


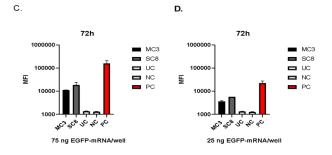
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Figures

E.





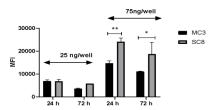


Figure 1. Graph of Mean Fluorescence Intensity (MFI) of EGFP-mRNA LNPs at different concentrations and times (A and B) Graph after 24 hours with 75 ng (A) and 25 ng (B). (C and D) Graph after 72 hours with 75 ng (C) and 25 ng (D) MFI summary of MC3 vs SC8 (E). Abbreviations: PC—positive controls, NC—negative controls, UC—untreated cells