Microfluidic-Driven Precision Lipid Nanoparticles for Nucleic Acid Delivery

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Lipid nanoparticles (LNPs) are a safe and effective non-viral platform for nucleic acid delivery, with broad applications in genome editing and RNA-based therapies [1,2]. This study describes the microfluidic-assisted formulation and optimization of LNPs tailored for two distinct therapeutic strategies: plasmid DNA delivery for genome editing and siRNA/miRNA delivery for the treatment of diabetic nephropathy (DN). For genome editing, LNPs encapsulating the AAVS1-CAG-eGFP plasmid were formulated using defined molar ratios (ionizable lipids: helper lipids: cholesterol: PEGylated lipids = 35:35:27.5:2.5) and tested across varying concentrations and helper lipid compositions. The optimal formulation (10 mM total lipid concentration, DSPC-based) exhibited well-controlled particle size, low polydispersity index, and high transfection efficiency (up to 90% in HeLa cells), with minimal cytotoxicity. For DN therapy, LNPs were designed to deliver siRNA targeting UBE2v1 and a miRNA mimic (hsa-miR-27b-3p), which modulate key pathogenic pathways in HK-2 renal cells [3]. These LNPs, formulated with a distinct lipid composition (ionizable lipids: helper lipids: cholesterol: PEGylated lipids = 50:10:1.5:38.5), demonstrated efficient cytoplasmic uptake, robust gene silencing, and excellent biocompatibility under hyperglycemic conditions. Current in vivo studies are focused on optimizing delivery routes and evaluating the tissue distribution of both LNP systems in mouse models. These findings underscore the transformative potential of microfluidic-engineered LNPs as a versatile and safe platform for targeted nucleic acid delivery, supporting the advancement of precision genome editing and novel RNA-based therapies across diverse therapeutic applications.

References

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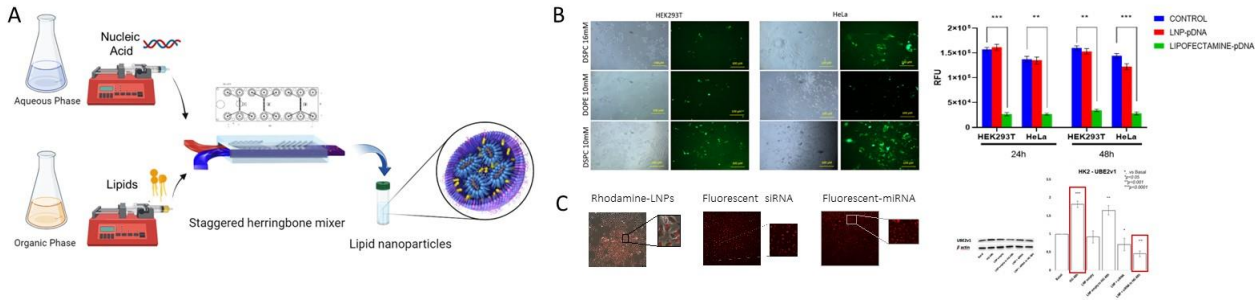

Figure 1: A) Schematic representation of the microfluidic formulation process of LNPs using a staggered herringbone mixer. B) Fluorescence microscopy images of HEK293T and HeLa cells comparing transfection efficiencies between plasmid DNA-loaded LNPs and Lipofectamine 3000, alongside cell viability assays post-transfection. C) Fluorescence microscopy images showing uptake of Rhodamine-labelled LNPs and fluorescently labelled siRNA and miRNA mimics in HK-2 human renal cells, with accompanying western blot results demonstrating UBE2V1 gene silencing by custom siRNA and downregulation of UBE2V1 expression via overexpression of custom miRNA mimics.