Impact of Nucleic Acid Backbone Modifications on the Morphology of Lipid Nanoparticles

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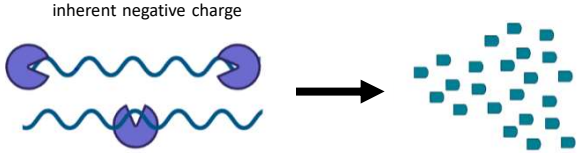
The Problem

Barriers to Gene Therapy

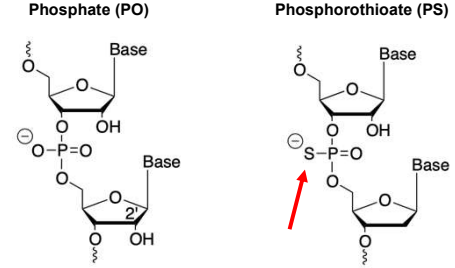
- Nucleic acid therapeutics are subject to nuclease degradation and an inability to cross cell membranes efficiently due to their inherent negative charge



The Solution

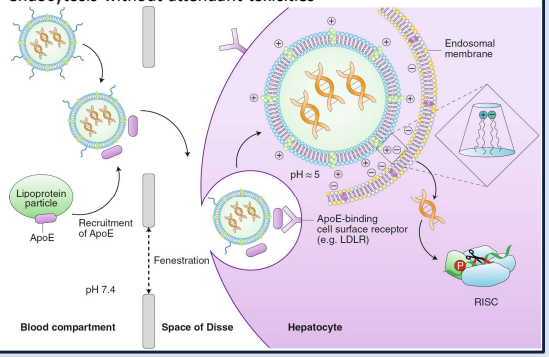
Improvements to the Construct

- Phosphorothioate backbone modifications in the nucleic acid sequence exert protection against nucleases



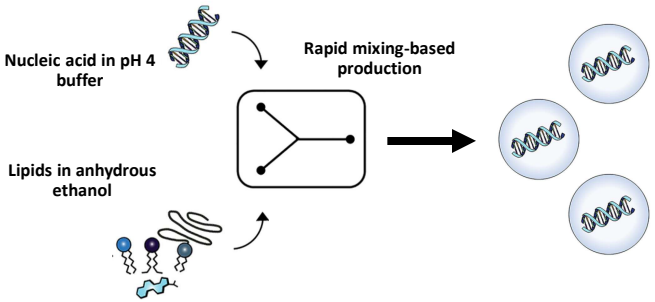
Improvements to Delivery

- Lipid nanoparticles enable nucleic acid encapsulation and intracellular delivery through ApoE-dependent receptor-mediated endocytosis without attendant toxicities



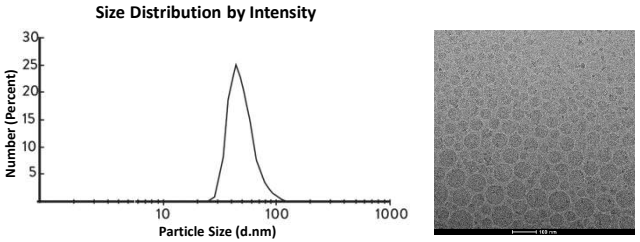
Our Methodology

Lipid Nanoparticle Formulation

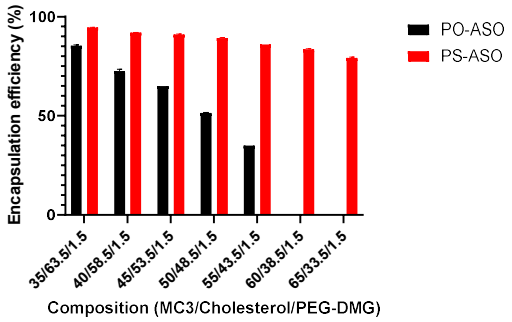


Structural Determination

- Particle size characterized by dynamic light scattering and morphology by cryogenic transmission electron microscopy

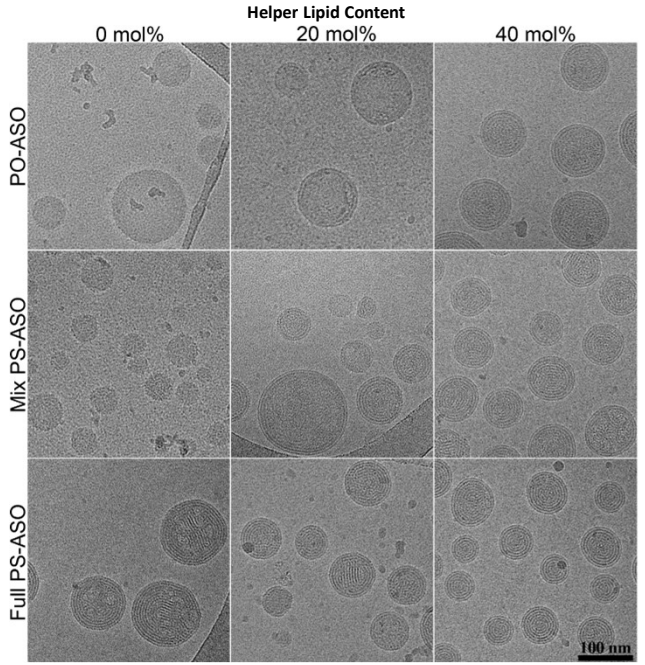


Phosphorothioate Modifications Enhance Entrapment



Phosphorothioate Modifications Alter Morphology

- Unmodified antisense oligonucleotides (PO-ASO) display uniformly dense cores up to 20 mol% helper lipid
- Fully phosphorothioate modified antisense oligonucleotides (fullPS-ASO) display striated internal structures across all compositions
- Antisense oligonucleotides with alternating phosphorothioate modifications (mixPS-ASO) display an in-between profile



Conclusions and Future Directions

- Phosphorothioate modifications impact LNP morphology substantially due to stronger interactions with ionizable amino lipids, resulting in enhanced entrapment
- Assessing the importance of modification localization as well as the minimum number of modifications required for maximal entrapment will further explain said phenomenon

Acknowledgements

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