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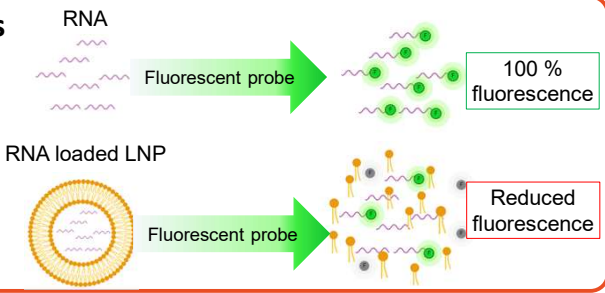
# Overcoming Lipid-Induced Quenching in RNA Quantification Using RiboGreen® Assay

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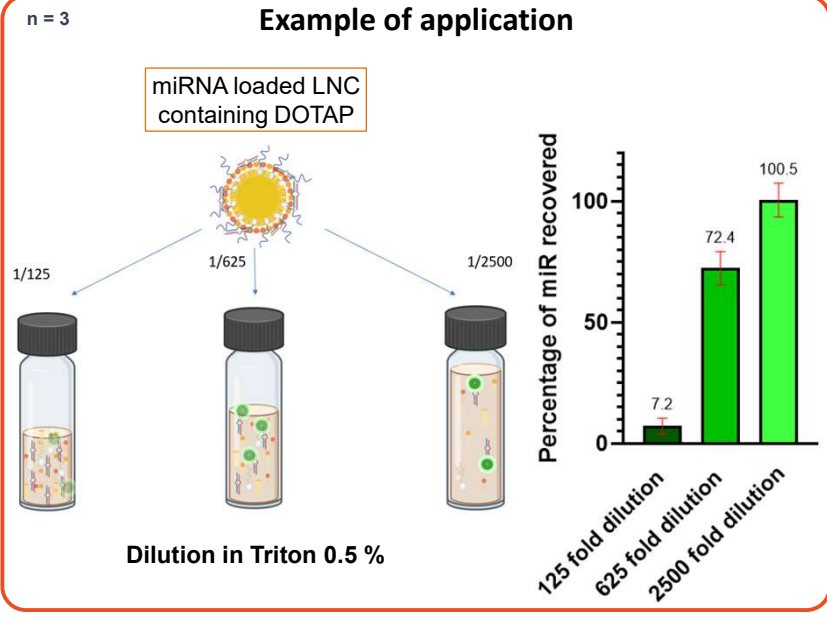
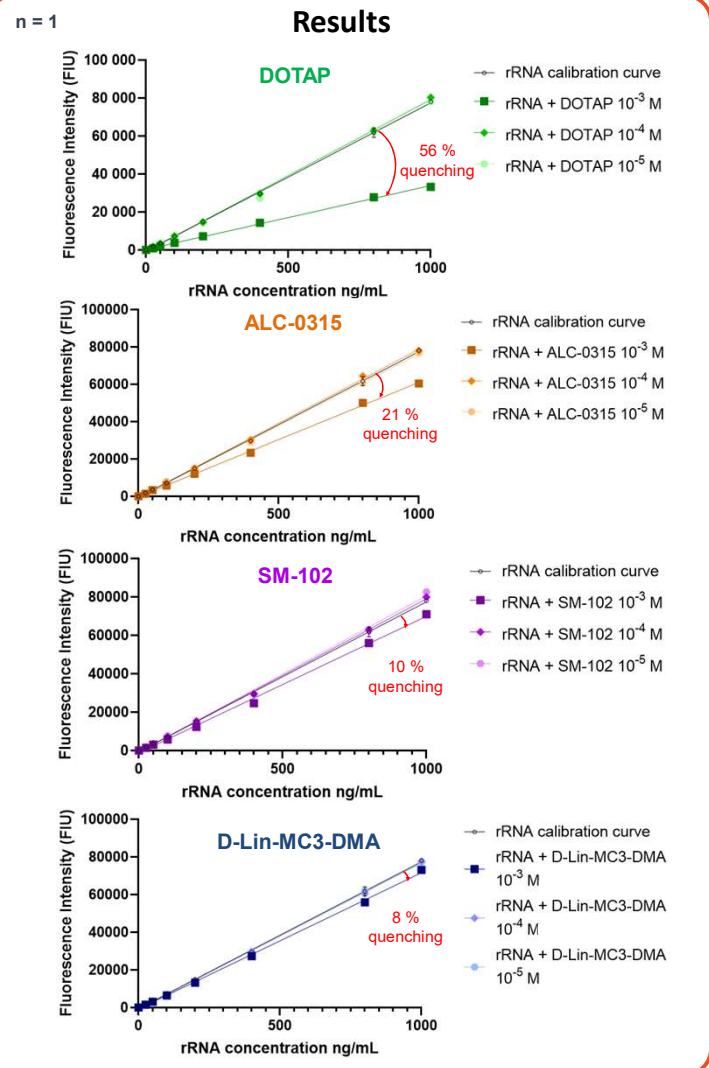
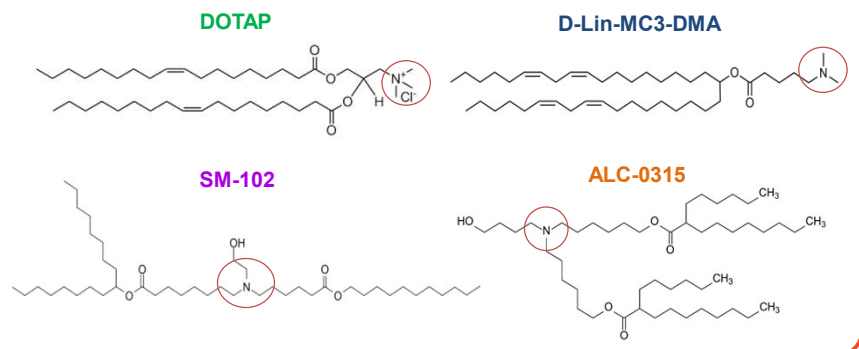
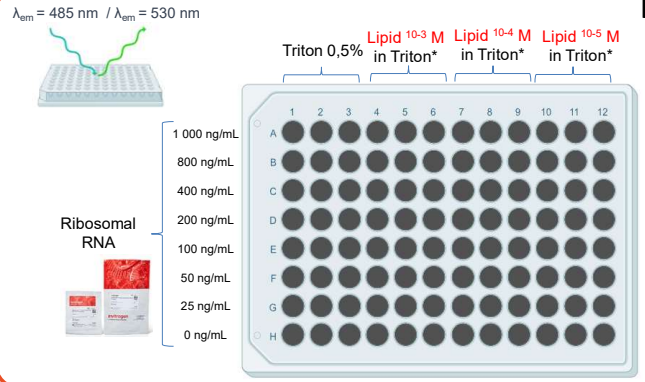
## Introduction & Objectives

Lipids used in lipid nanoparticles (LNPs) leverage their ability to **complex** with nucleic acids, such as RNA, for **gene delivery applications**. Quantifying the RNA encapsulated in these nanoparticles presents a challenge due to **interference** from the lipids themselves. Among the common methods, the **RiboGreen®** assay is frequently used for **RNA quantification**, although most approaches are indirect. However, accurate quantification requires direct measurement of RNA concentration, while overcoming the quenching effect caused by the lipids.



**Objectives**  
This study aims to **evaluate** the lipid concentrations at which **quenching** becomes **significant** and to **determine the dilution factors** that can mitigate or eliminate this effect, ultimately **improving** the precision of **RNA quantification** in LNPs.

## Material & Methods



## Conclusion & Discussion

The lipids tested induce a **quenching** effect on RiboGreen® kit fluorescence at **high concentrations**, with DOTAP and ALC-0315 having the greatest impact (up to 56% and 21% reduction). This phenomenon seems to be **eliminated** by dilution regardless of the lipid. Appropriate dilution (up to 2500-fold,  $10^{-5} \text{ M}$ ) has enabled us to reliably quantify the **RNA** present in our Lipid Nanocapsules. Controlled dilution of lipid-containing samples in surfactant **eliminates the quenching effect** and ensures **accurate RNA quantification**. This optimization is essential for reliable assessment of encapsulation efficiency and drug loading.

## Perspectives

