Nanomedicine and Nanoscale Delivery

High-throughput screening of lipid nanoparticles for lung delivery of mRNA

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CRS 2025 ANNUAL MEETING & EXPOSITION
PHILADELPHIA, PA
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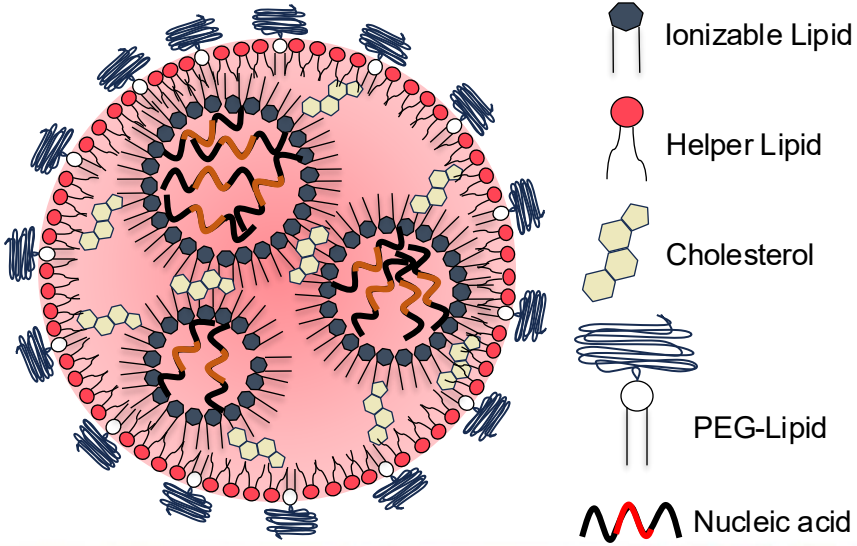
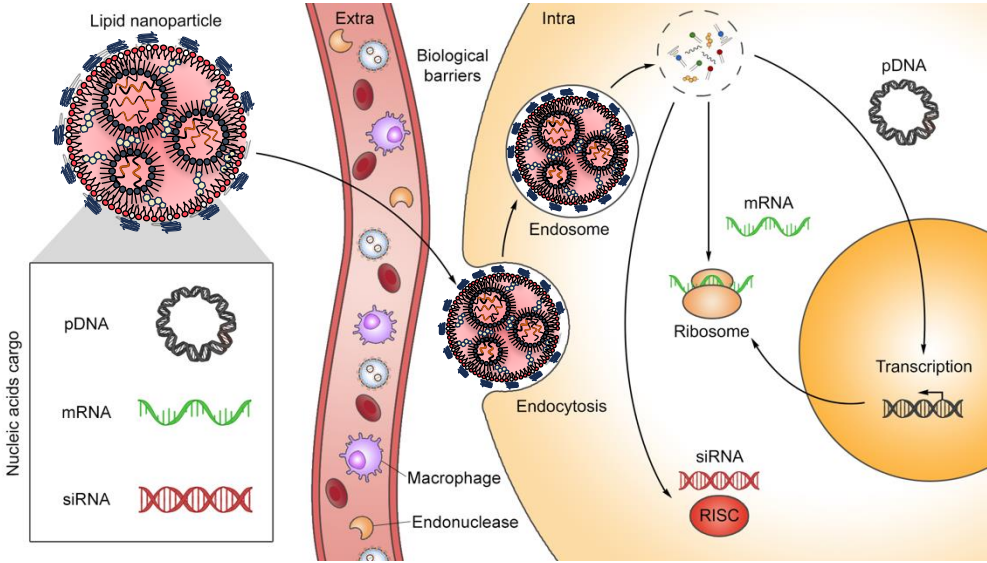
NEXT-GENERATION DELIVERY INNOVATIONS

INTRODUCTION

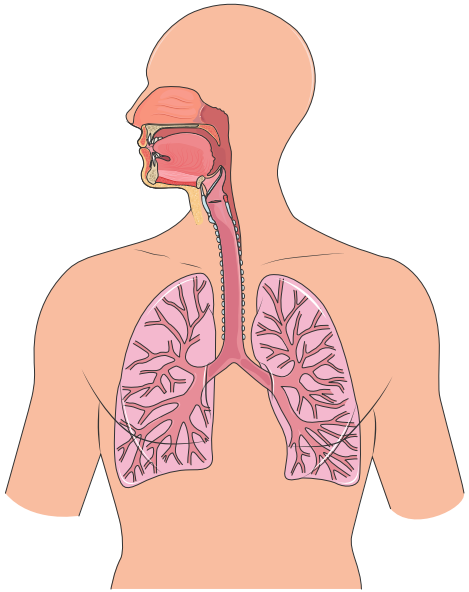
Ionizable Lipid Nanoparticles (LNPs)

1. Encapsulate nucleic acids
2. Prevent nucleic acid enzymatic degradation
3. Mediate intracellular delivery





Intranasal administration



Advantages:

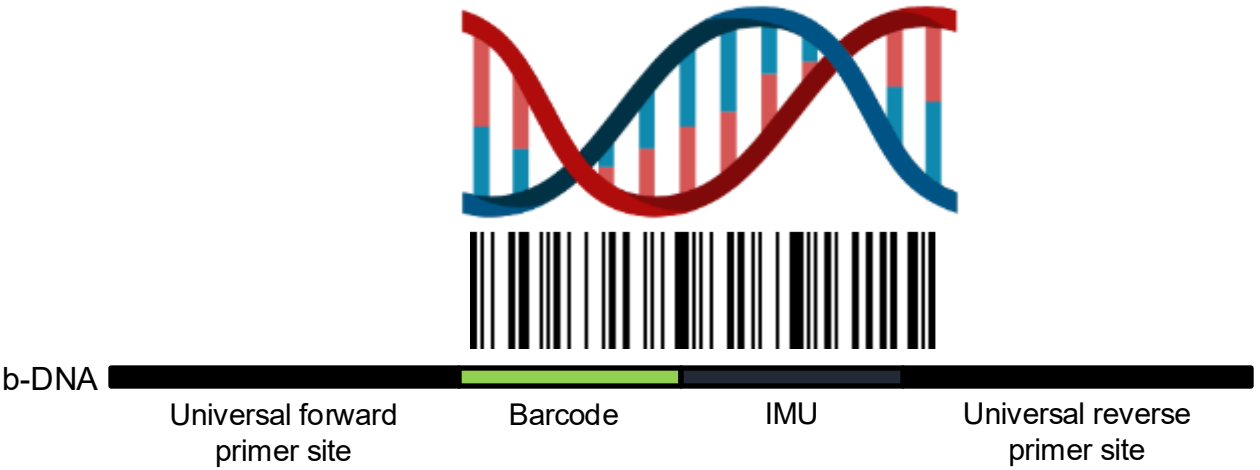
- ✓ Direct access to the lungs
- ✓ Reduced systemic exposure

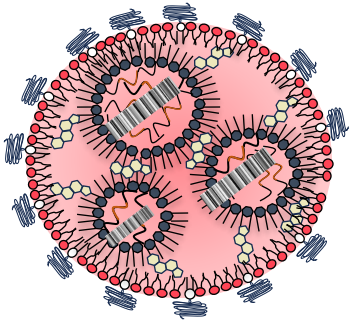
Challenges?

1. Mucus
2. Mucociliary clearance
3. Limited volume for administration
4. LNP composition
5. LNP physicochemical properties

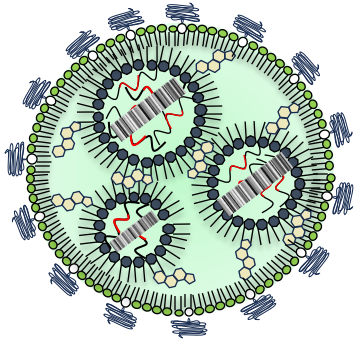
Barcode DNA (b-DNA) technology

Accelerates the identification of lead LNPs for targeted delivery, minimizing the number of animal used





LNP1
Barcode 1

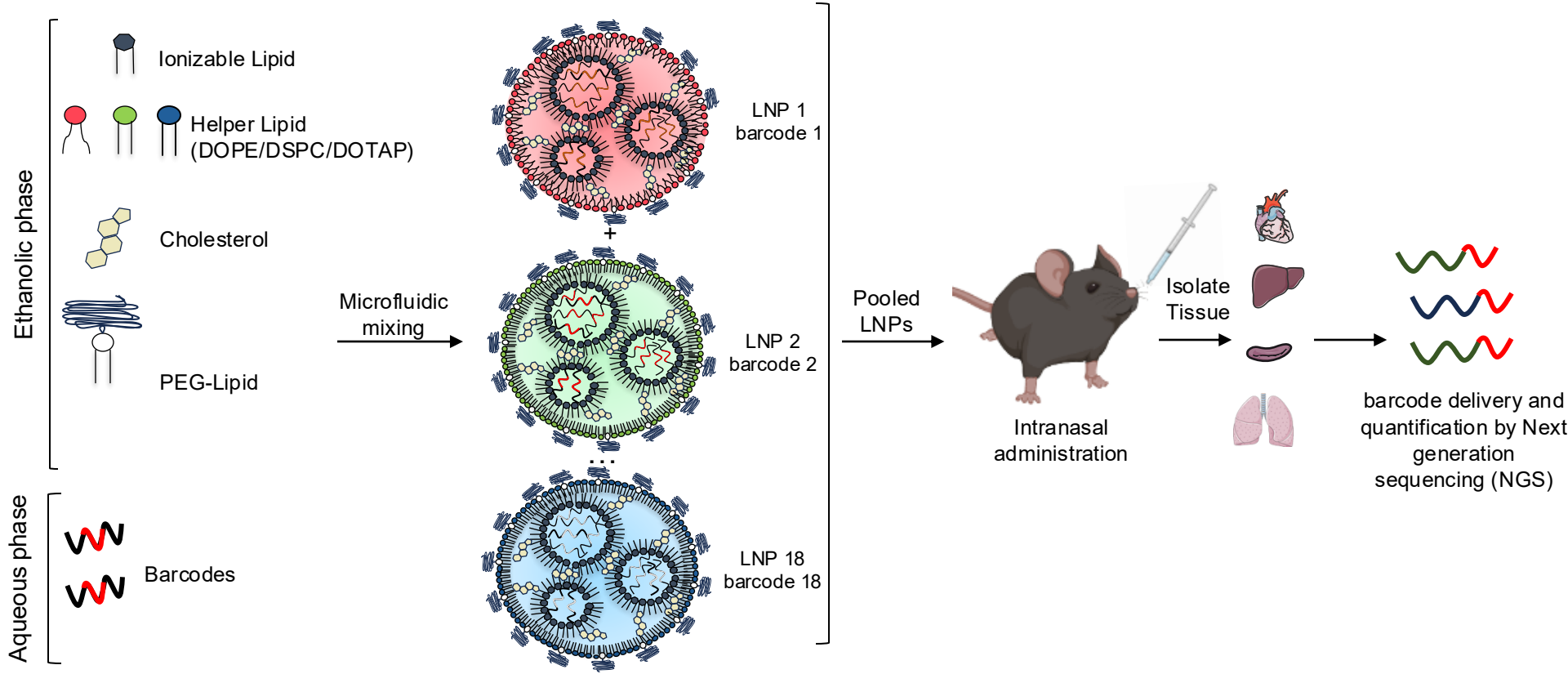


LNP2
Barcode 2

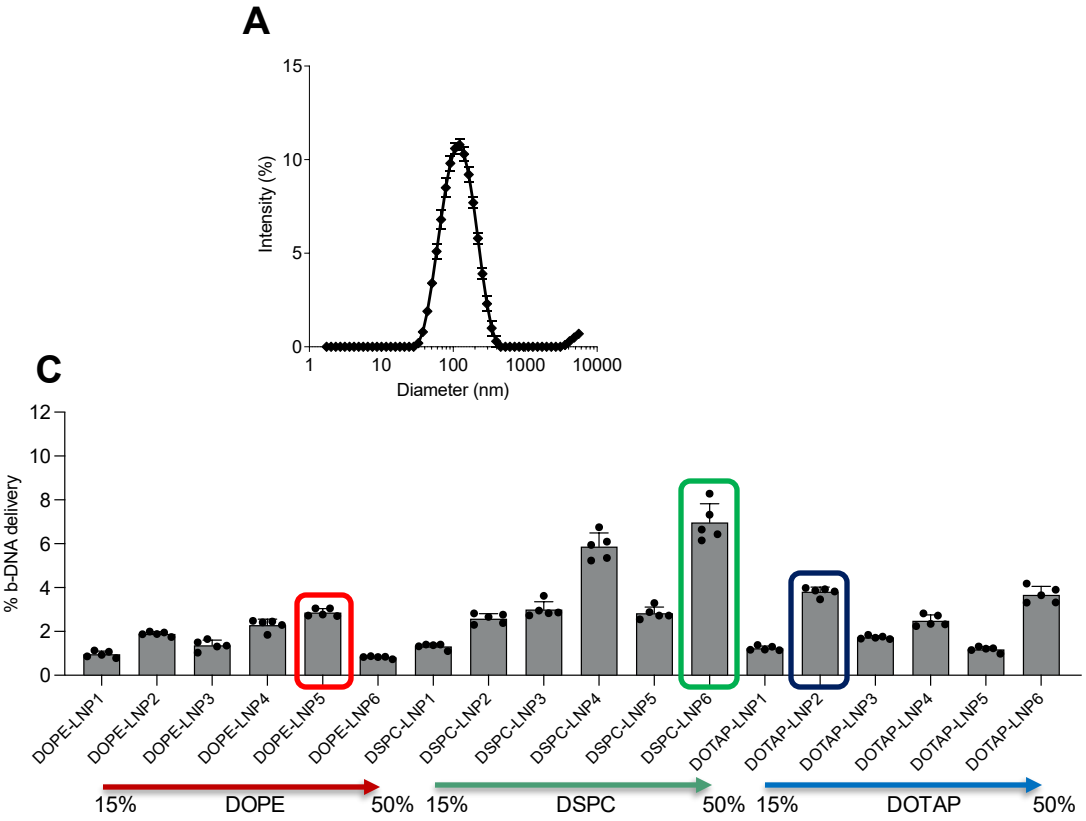
OBJECTIVE

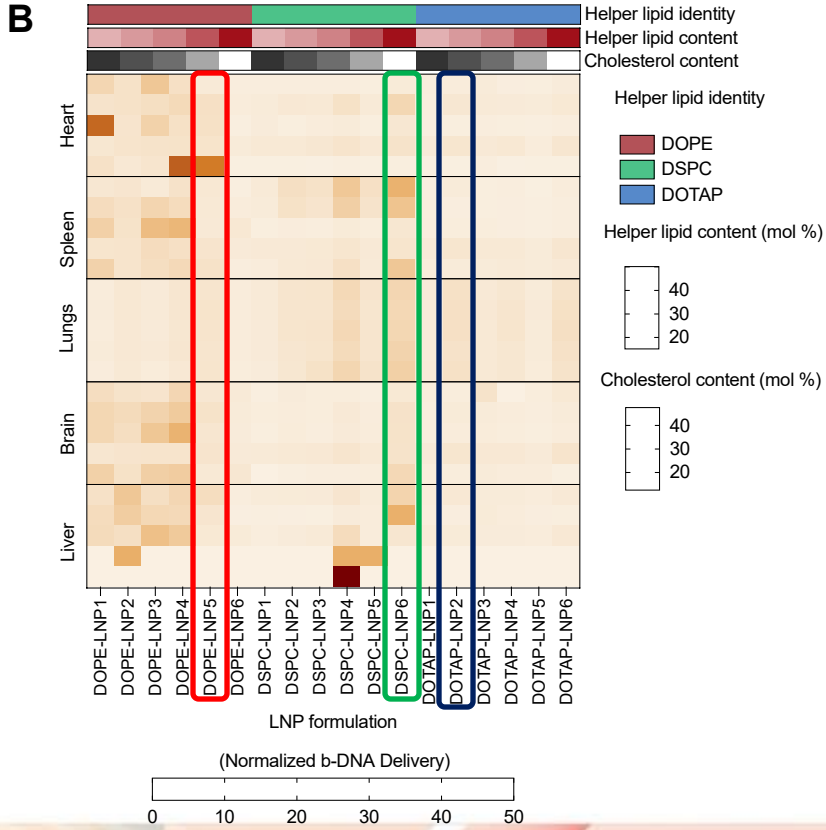
Identify an **LNP composition** able to **overcome** the **barriers of intranasal** administration and delivery **mRNA** to the **lungs**

High-throughput in vivo screening of barcoded-LNPs

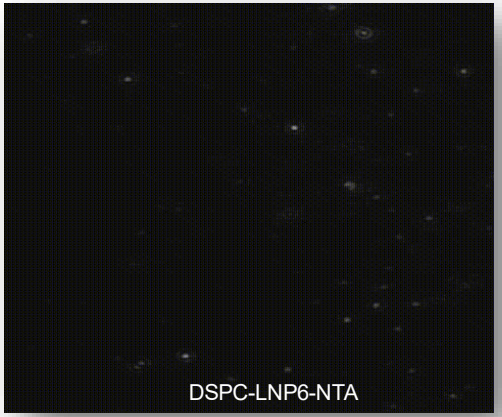


Barcoded LNPs screening

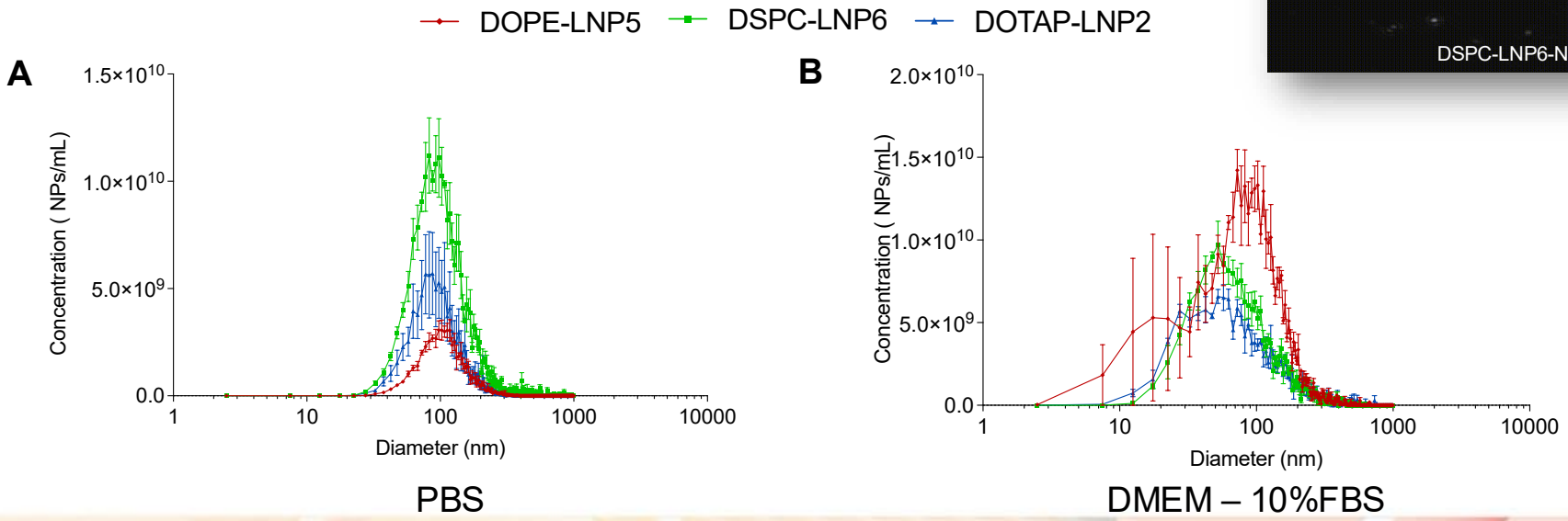




Physicochemical Characterization of LNPs in Cell Culture Medium



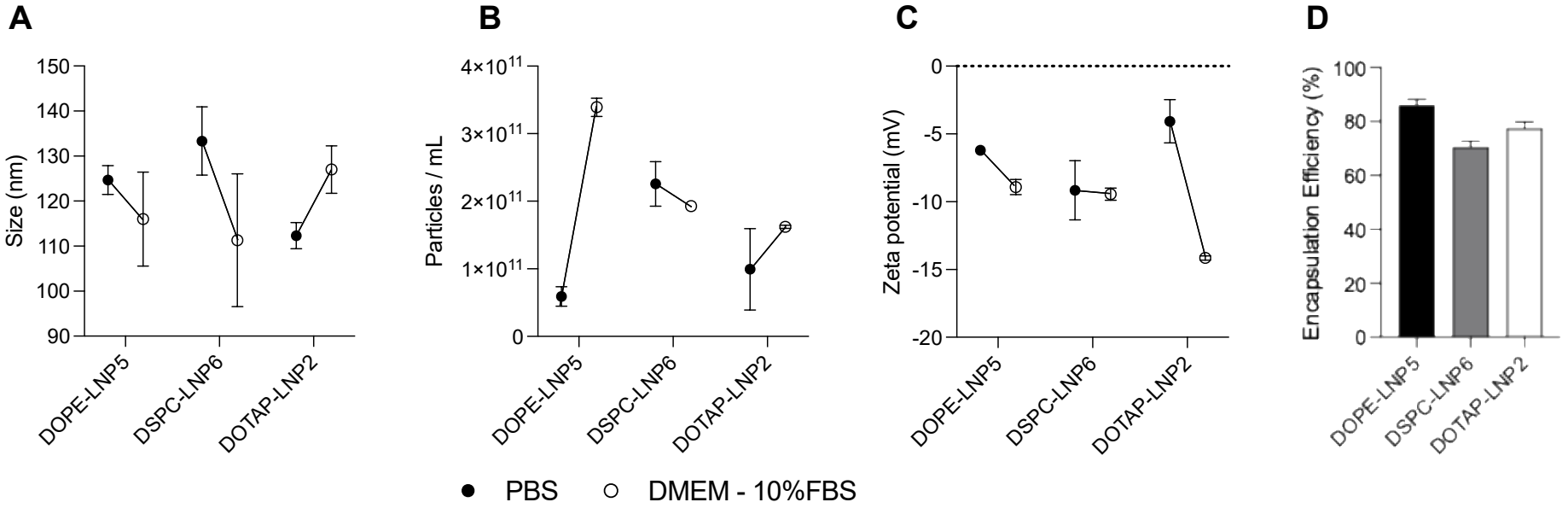
Nanoparticle tracker analysis (NTA)



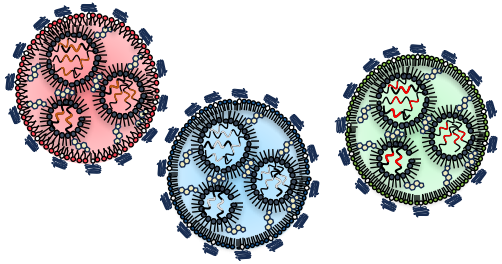
PBS

DMEM – 10%FBS

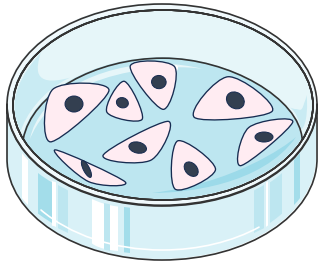
Physicochemical Characterization of LNPs in Cell Culture Medium and Encapsulation efficiency



in vitro transfection efficiency does NOT recapitulate b-DNA delivery to the lungs



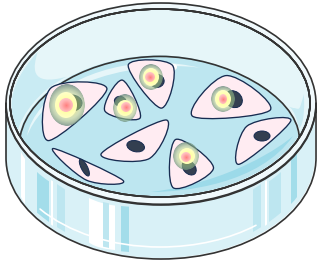
LNP treatment



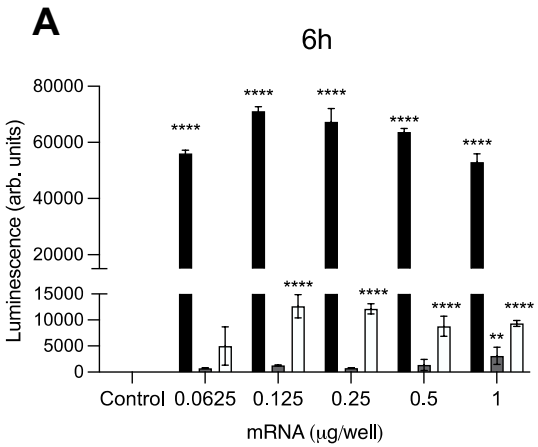
A549 Cell line

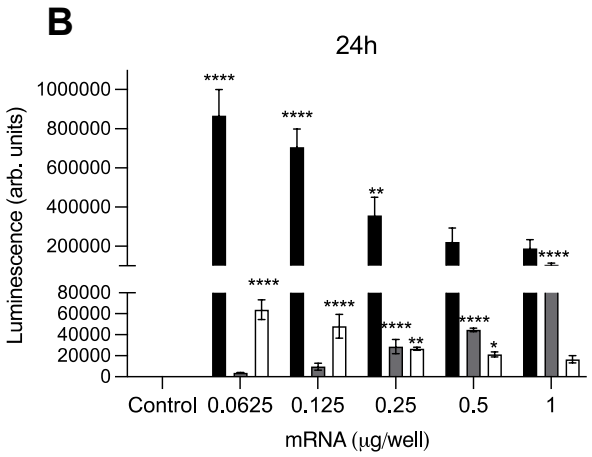
Transfection efficiency

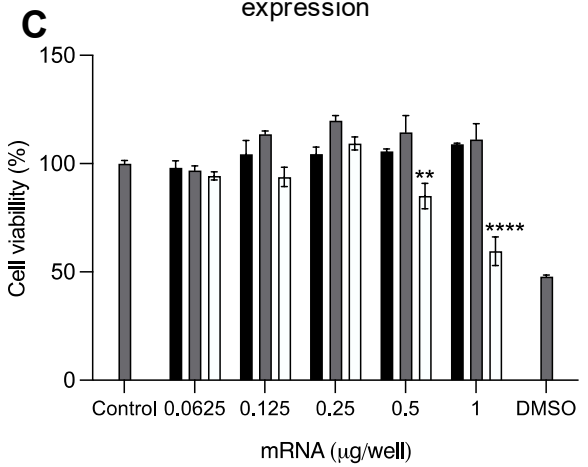
Cell viability



Luciferase expression

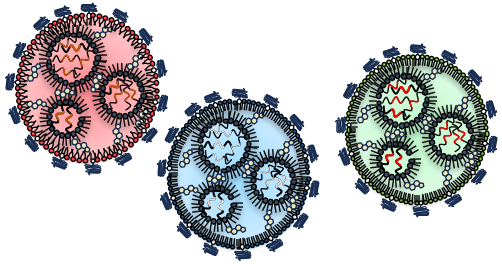




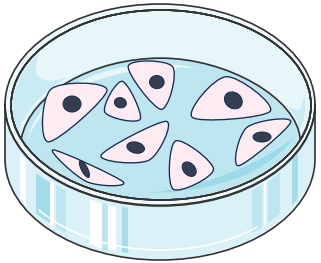


DOPE-LNP5 DSPC-LNP6 DOTAP-LNP2

What is the fairest comparison between LNPs?



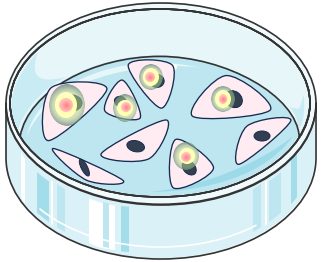
LNP treatment



A549 Cell line

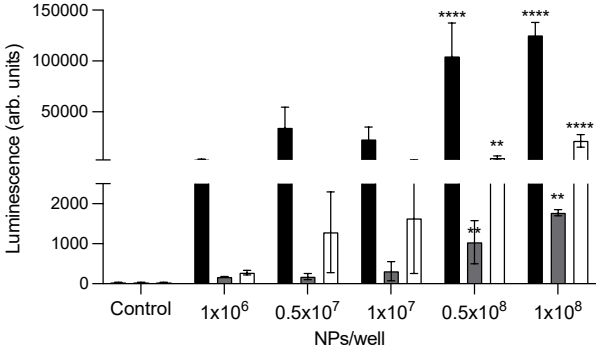
Transfection efficiency

Cell viability

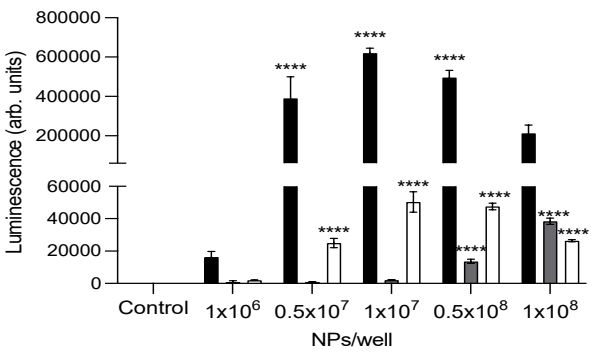


Luciferase expression

A



B



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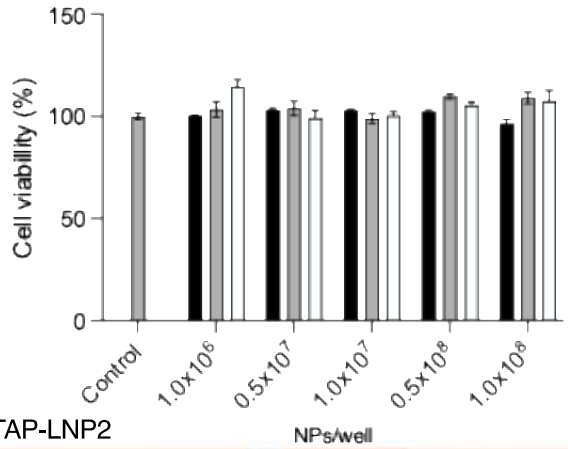
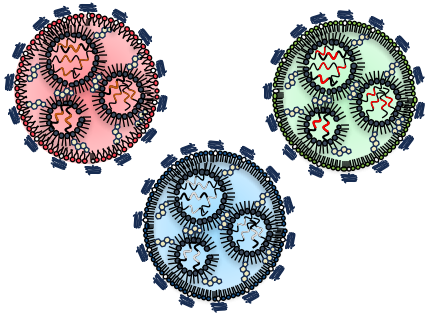


DOPE-LNP5 DSPC-LNP6 DOTAP-LNP2

In vivo transfection validation

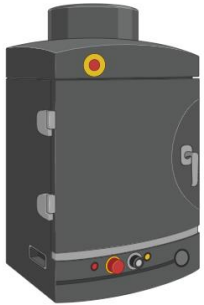


LNP treatment

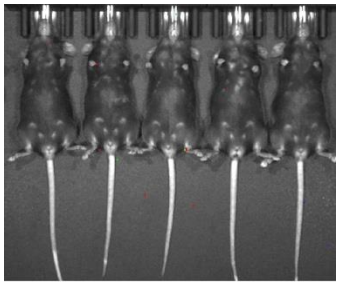
0.25 mg/kg of mRNA

4 h post-treatment

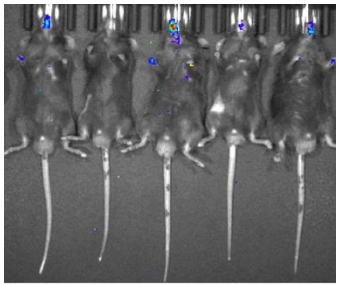
in vivo and *ex vivo* image acquisition



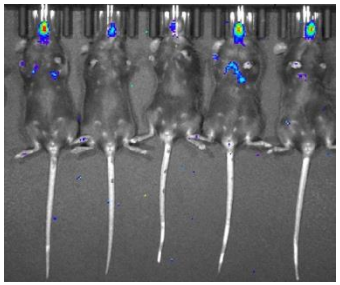
PBS



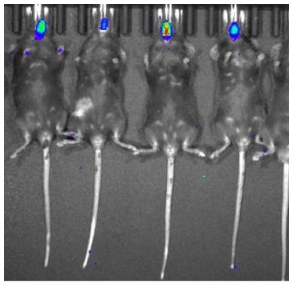
DOPE-LNP5

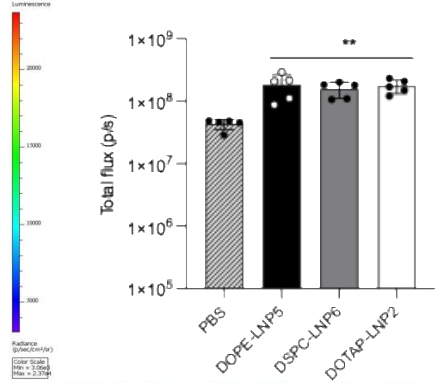


DSPC-LNP6

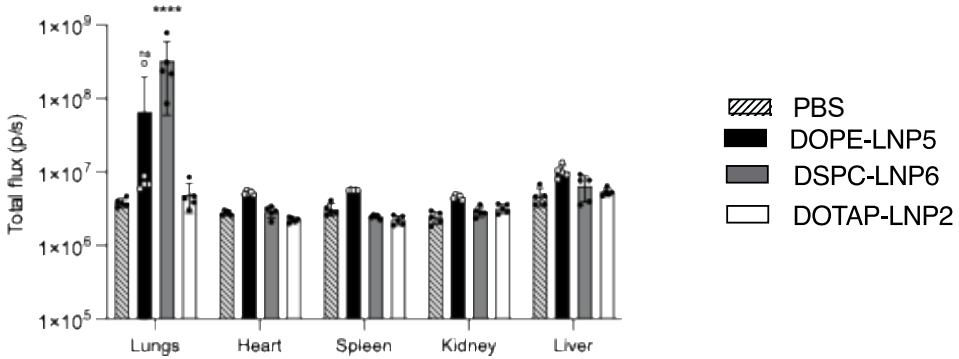


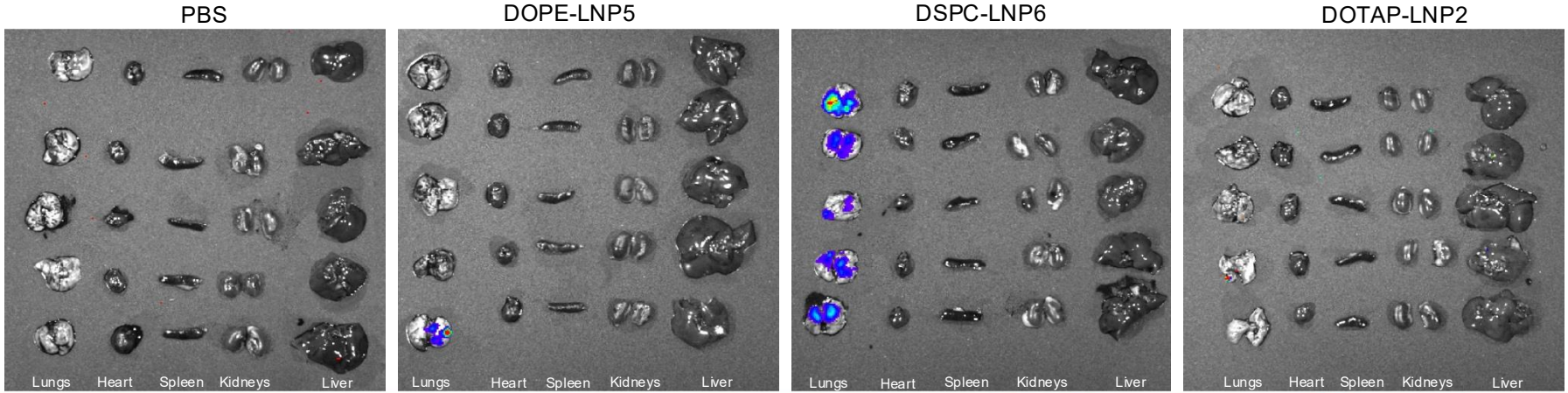
DOTAP-LNP2





In vivo transfection validation





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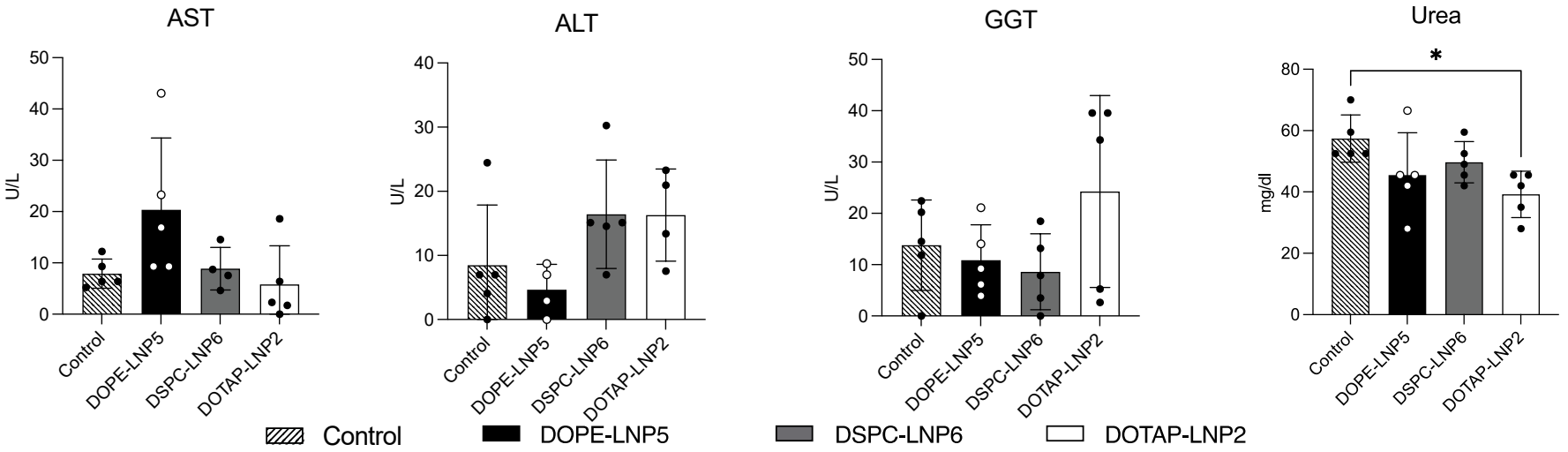
Safety profile – Hepatic and renal function after LNP administration



CONCLUSION

1. *Barcoded LNPs accelerate the identification of formulations for lung delivery via intranasal (IN) administration using a reduced number of animals.*

2. *Barcode screening successfully predicted in vivo efficacy.*

3. *No correlation was observed between barcode delivery and in vitro transfection in A549 cell line.*

4. *This screening strategy identifies DSPC-LNP6 as a promising candidate for gene therapy applications targeting lung diseases.*

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Laboratório de Biofísica de Sistemas Nanoestruturados

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THANKS

I’d be happy to take questions