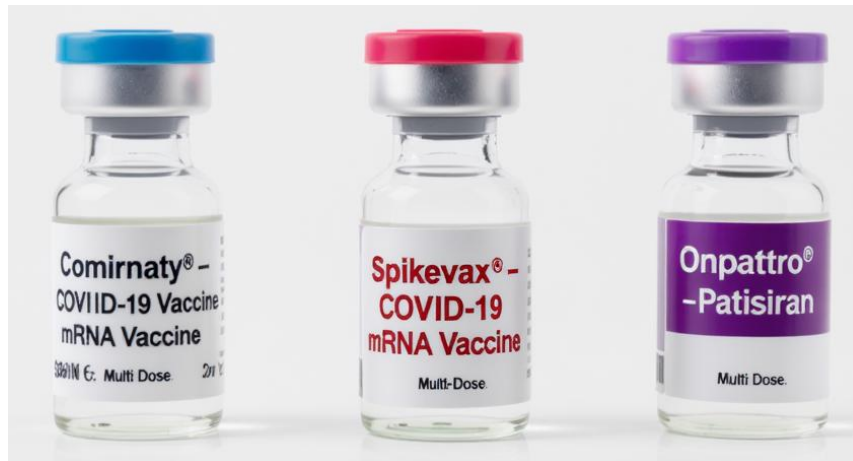
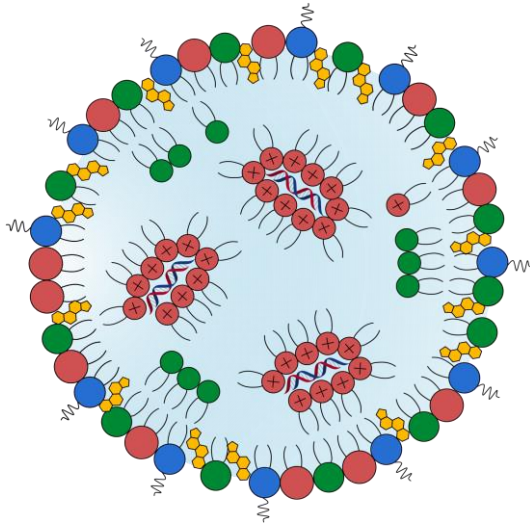


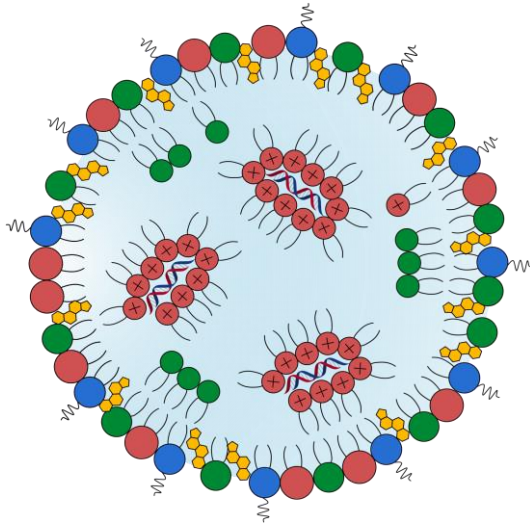
Dual Centrifugation - A Novel Perspective on LNP Formulation Development

Valentin Bender

LNP Formulation Development

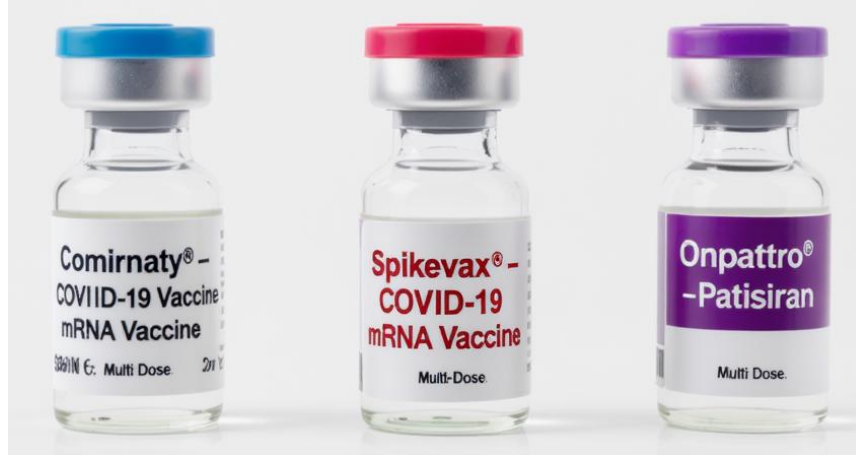


LNP Formulation Development

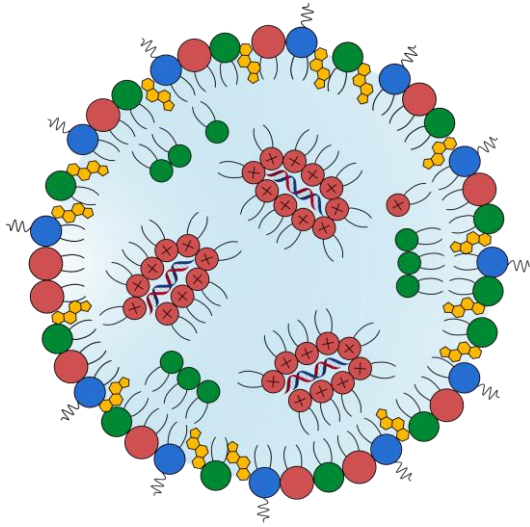


Drawbacks of established manufacturing processes

- Costs intensive
- High resource consumption
- Time consuming manufacturing techniques



LNP Formulation Development

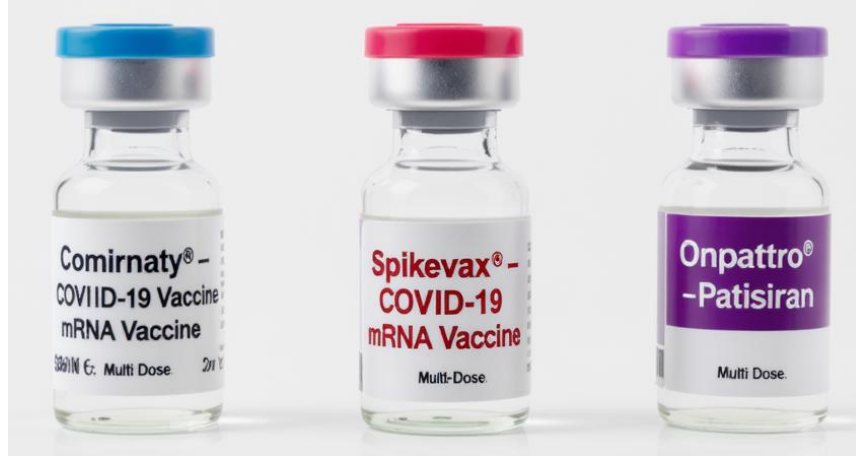


Drawbacks of established manufacturing processes

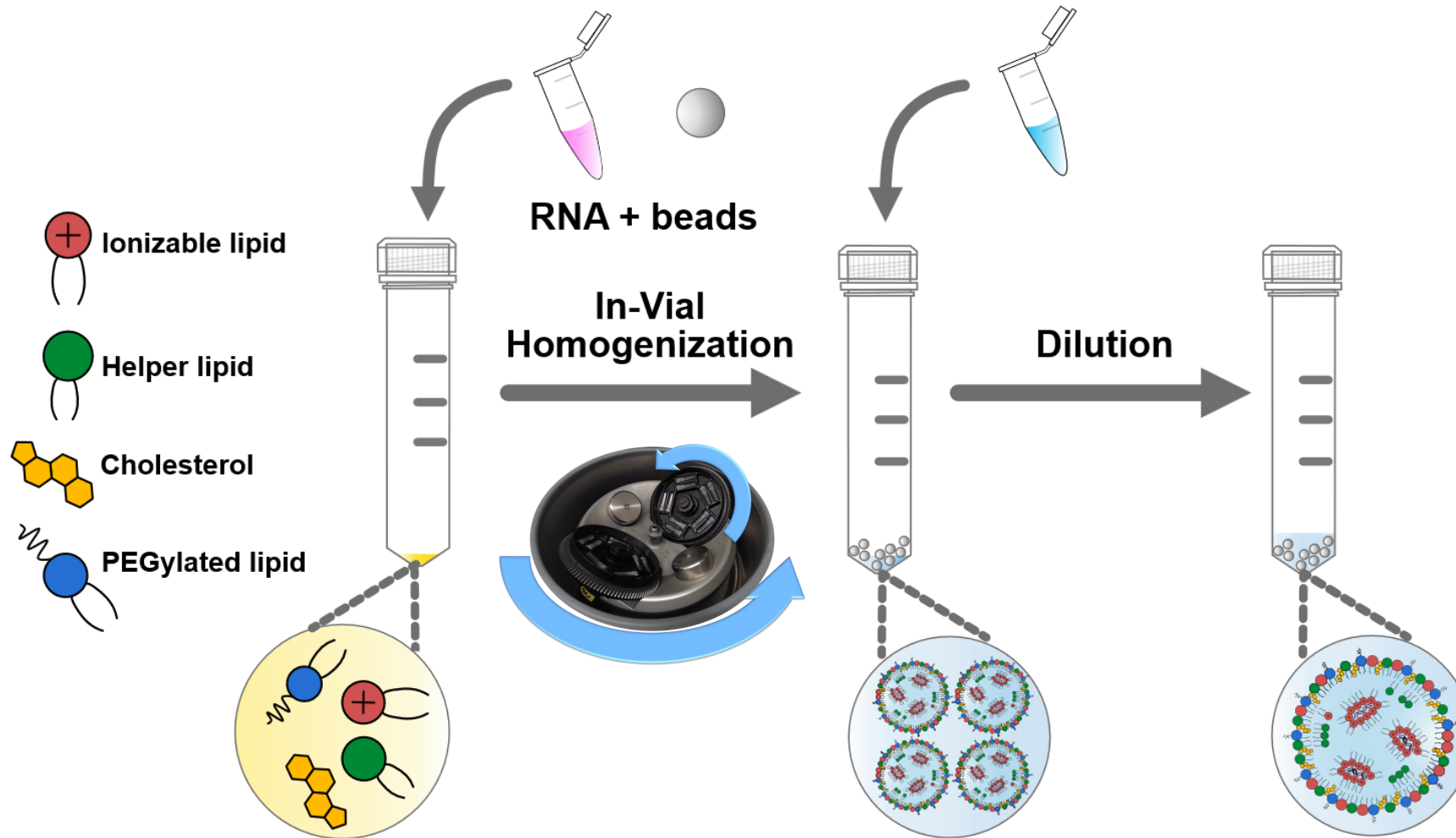
- Costs intensive
- High resource consumption
- Time consuming manufacturing techniques

Aims

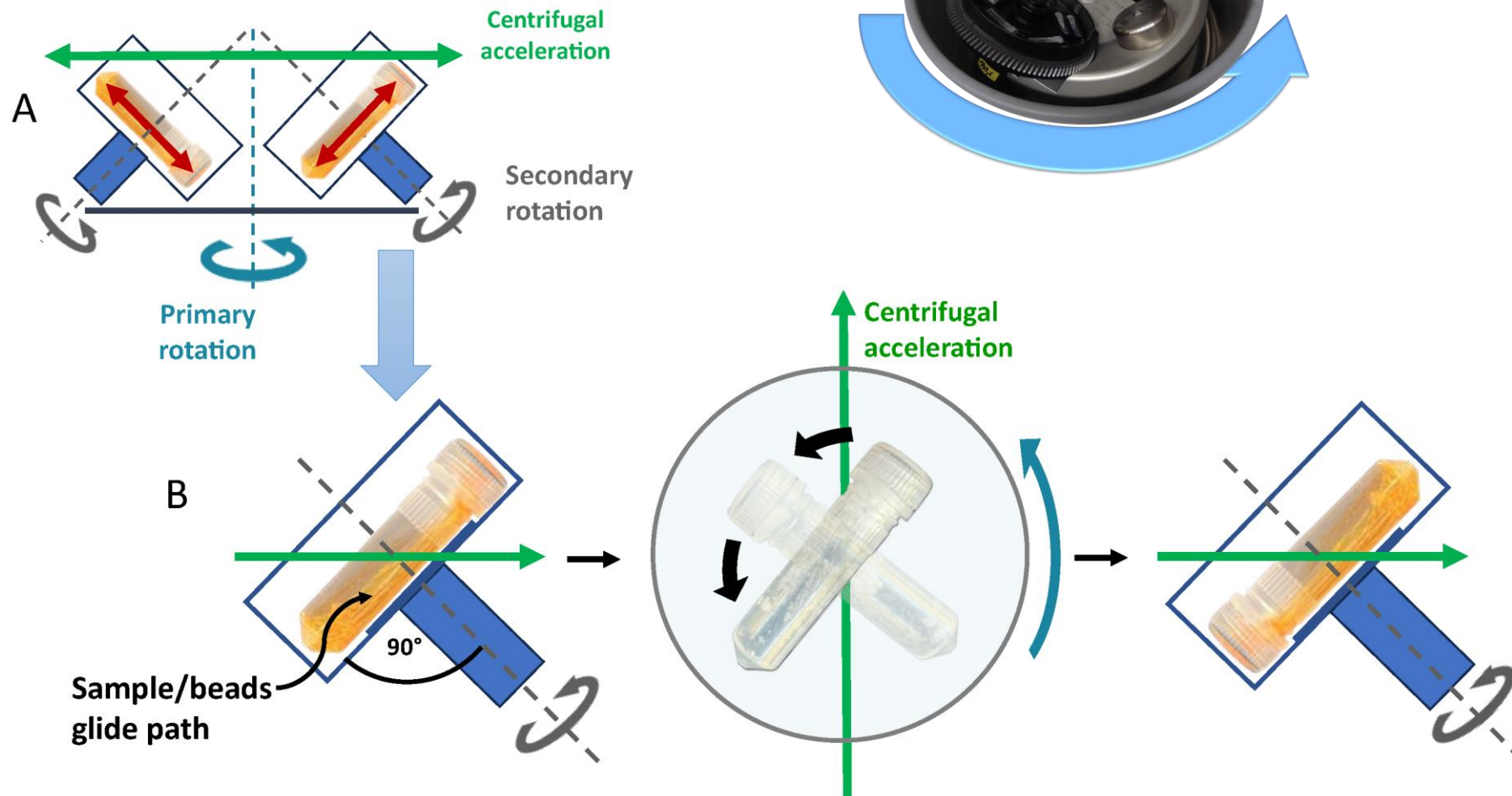
- Developing a fast and robust **high-throughput screening** platform
- Minimizing **resource consumption**
- Personalized therapeutic strategies via **bedside preparation**



LNP Preparation *via* In-Vial Homogenization



In-Vial Homogenization



Koehler, Schmager, Bender et al.,
Pharmaceuticals, (2023)
doi.org/10.3390/ph16111519

Experimental Setup

Process Parameters

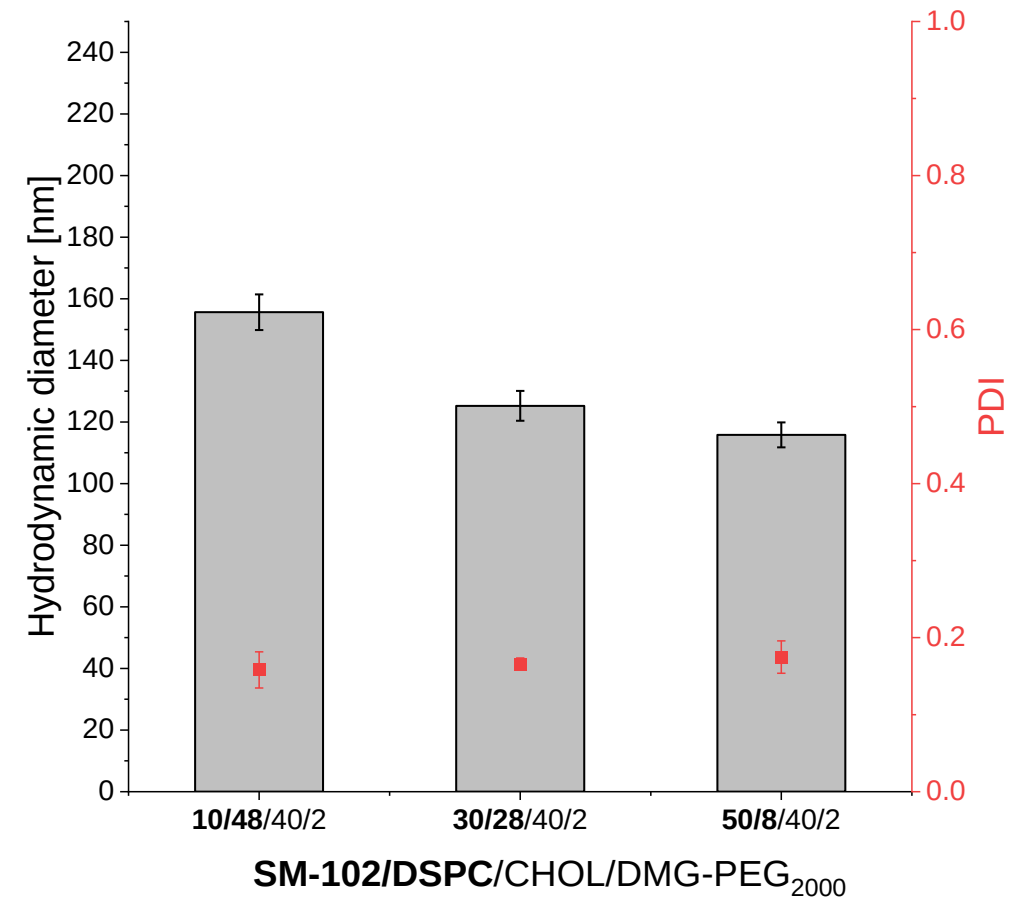
Gentle *in-vial* homogenization

Rapid formulation process → **Total 10 min**

Very small batches → **1 mg** lipid mass

LNP Characteristics

- miRNA payload
- N/P ratio of 6



Data shown as
mean $\pm$ SD (n=3)

Experimental Setup

Process Parameters

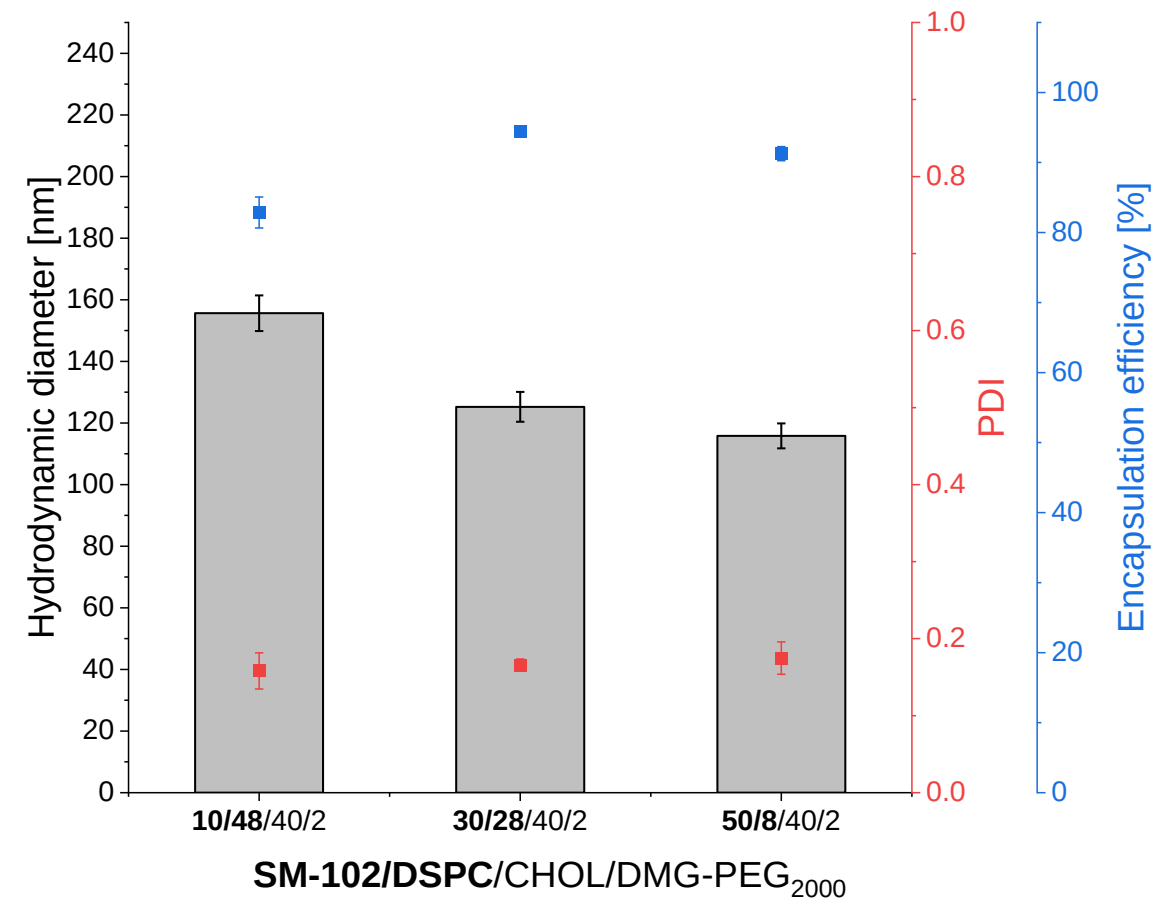
Gentle *in-vial* homogenization

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- miRNA payload
- N/P ratio of 6



Data shown as
mean ± SD (n=3)

Experimental Setup

Process Parameters

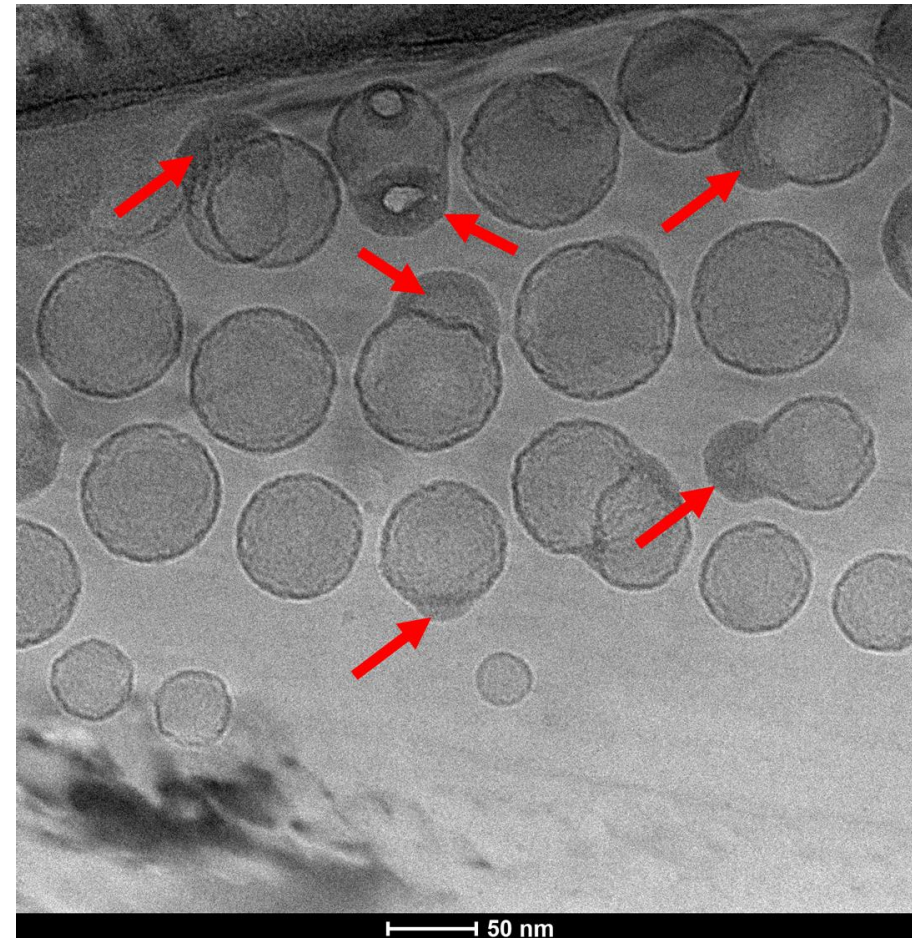
Gentle *in-vial* homogenization

Rapid formulation process → **Total 10 min**

Very **small batches** → **1 mg** lipid mass

LNP Characteristics

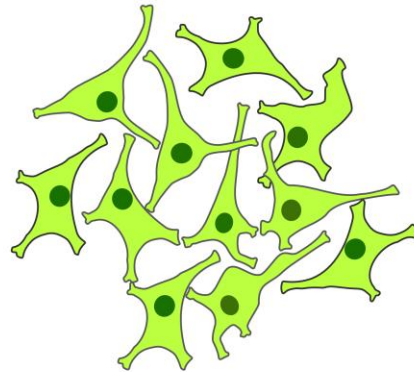
- miRNA payload
- N/P ratio of 6



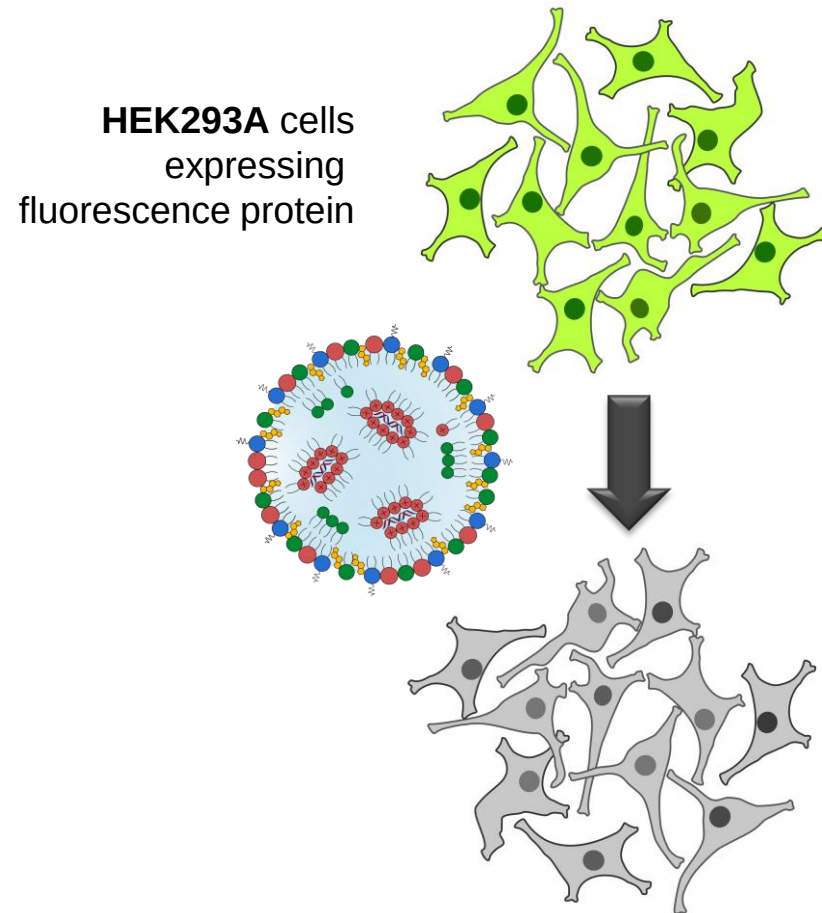
Data shown as
mean $\pm$ SD (n=3)

Knockdown Efficiencies

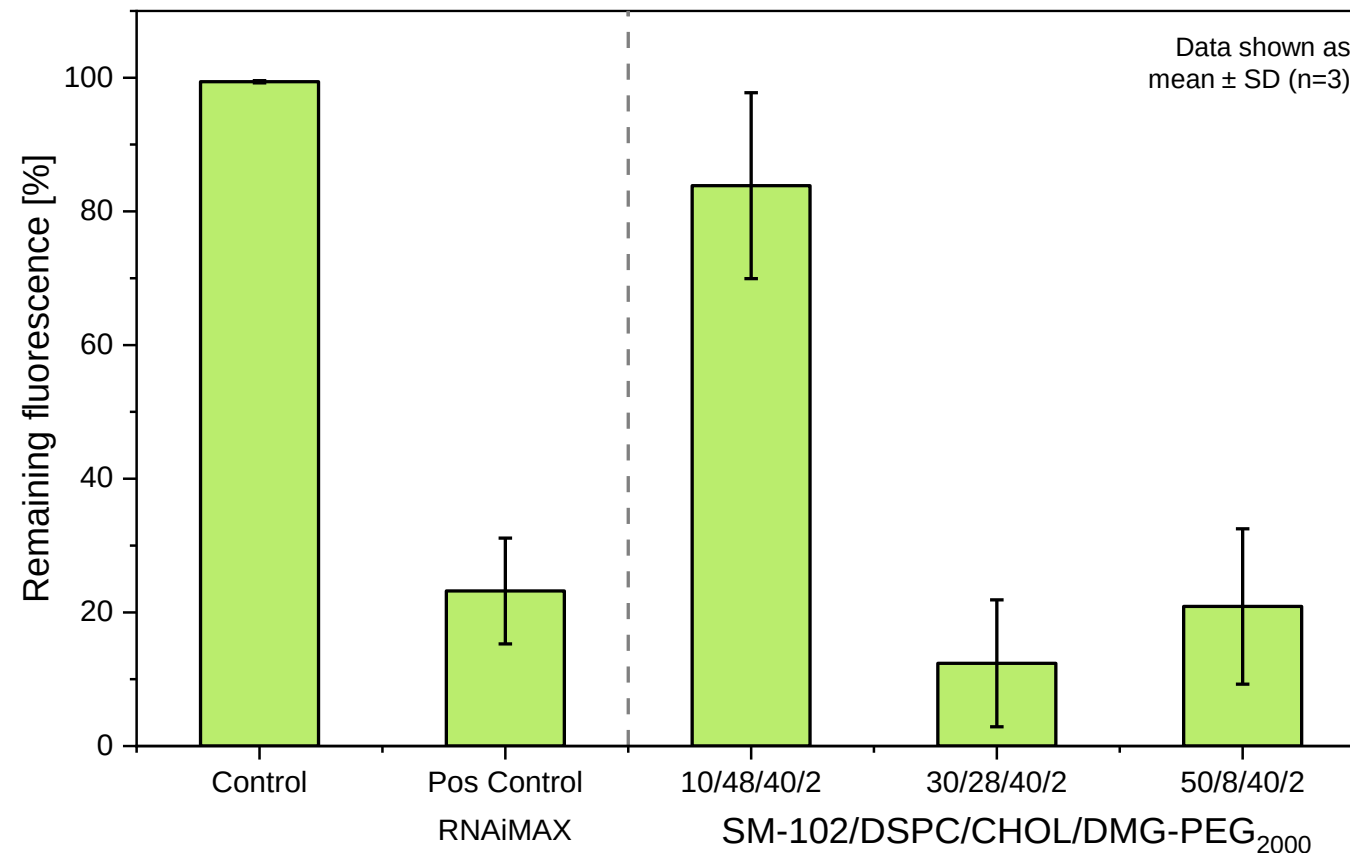
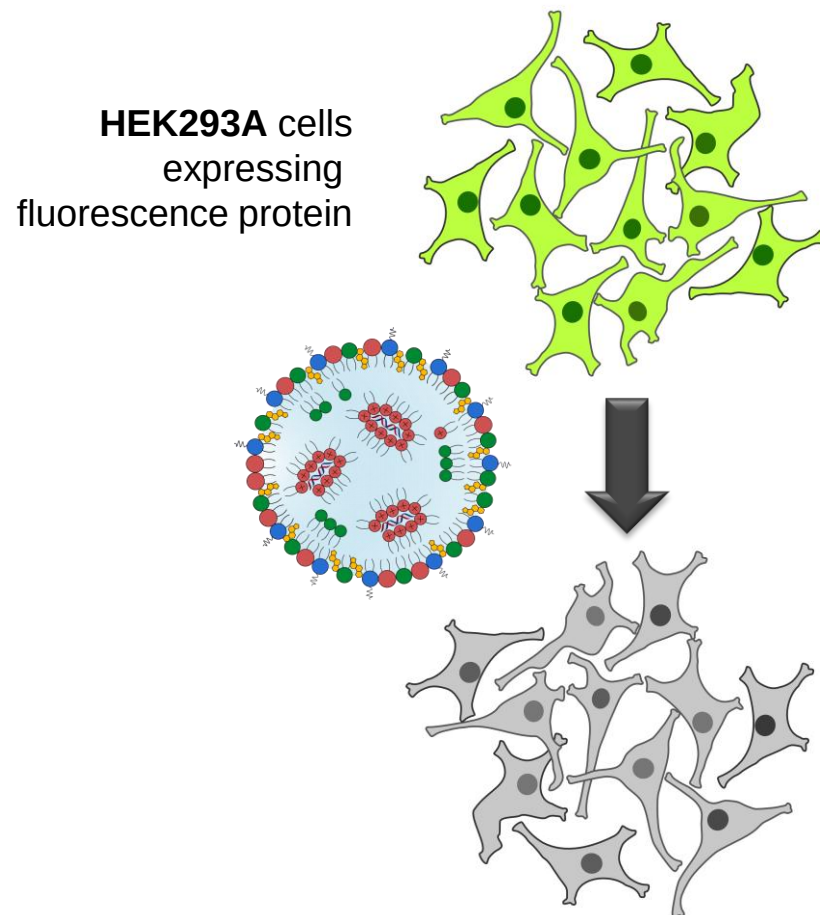
HEK293A cells
expressing
fluorescence protein



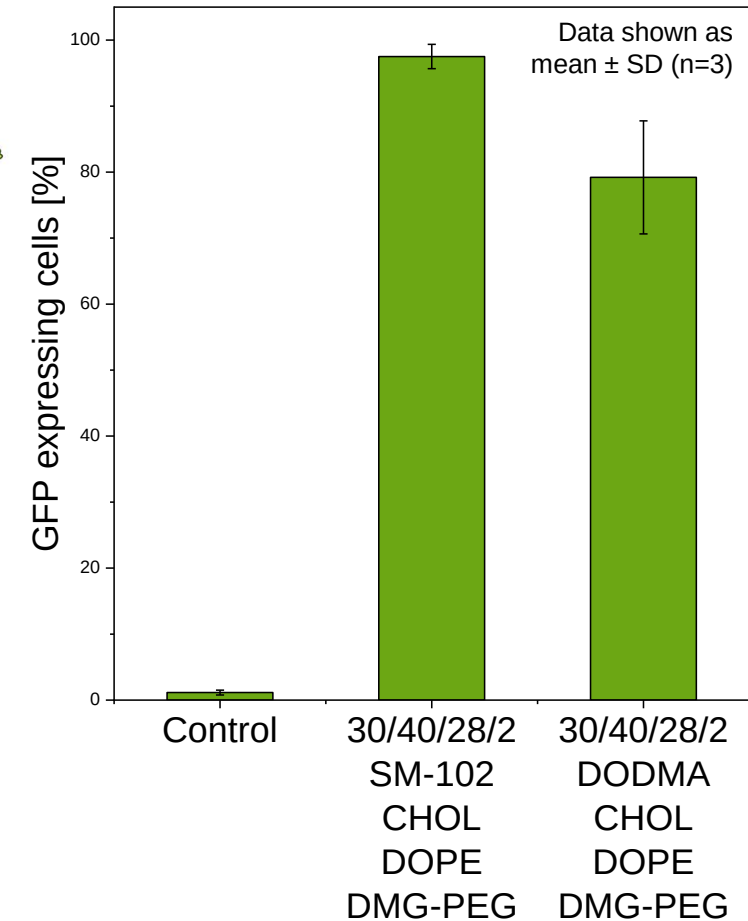
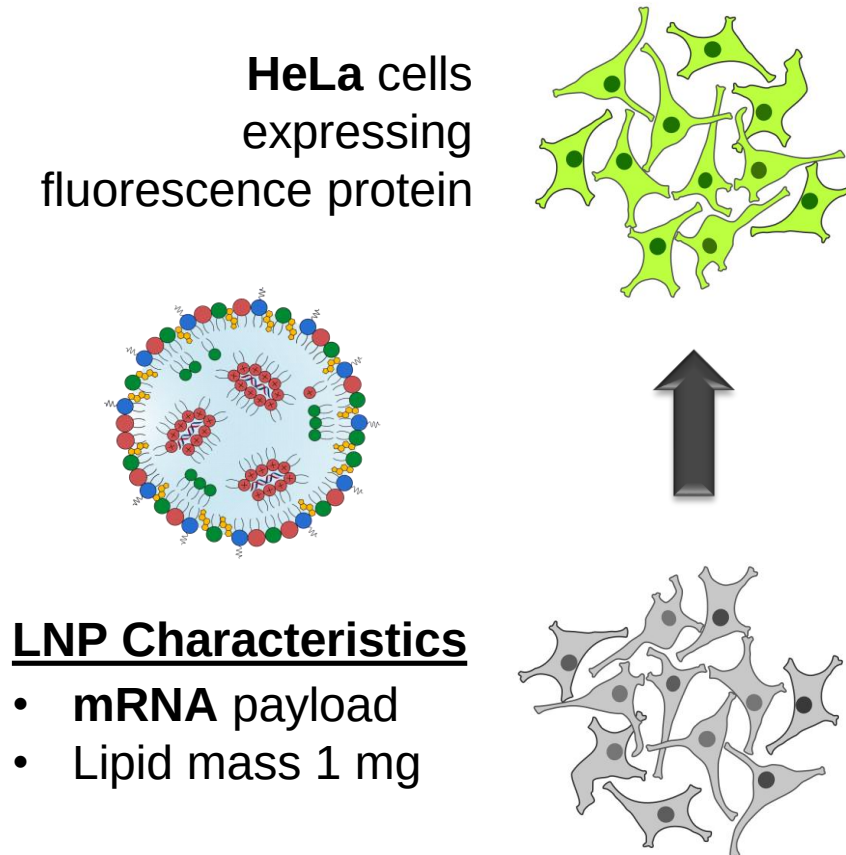
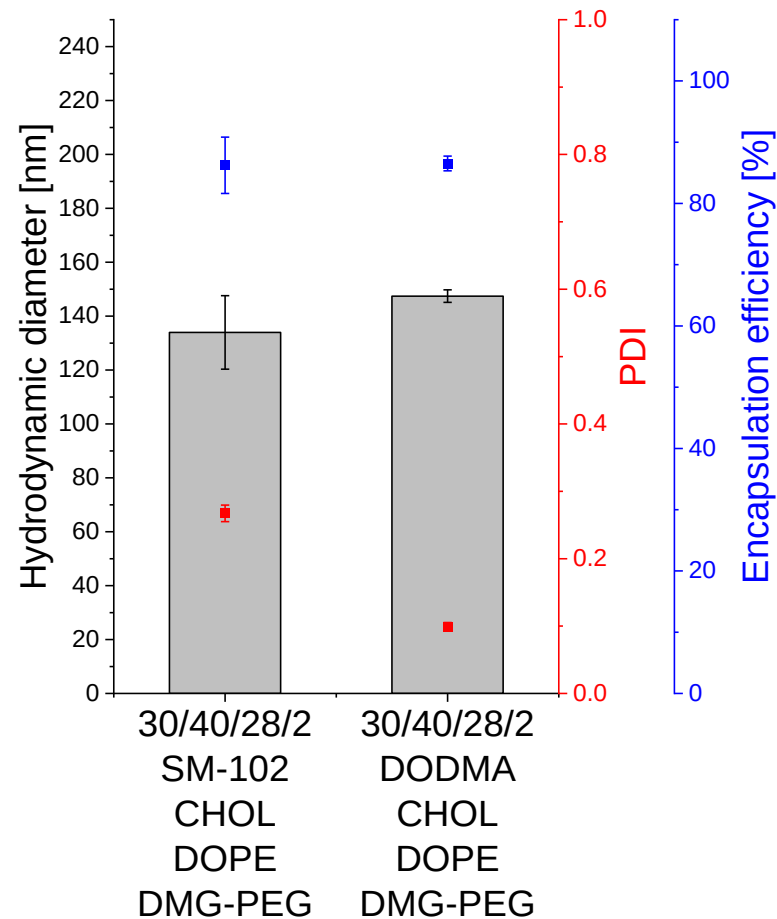
Knockdown Efficiencies



Knockdown Efficiencies

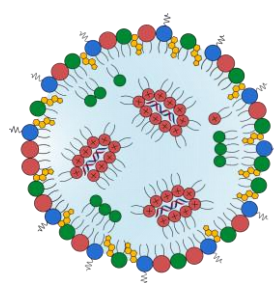
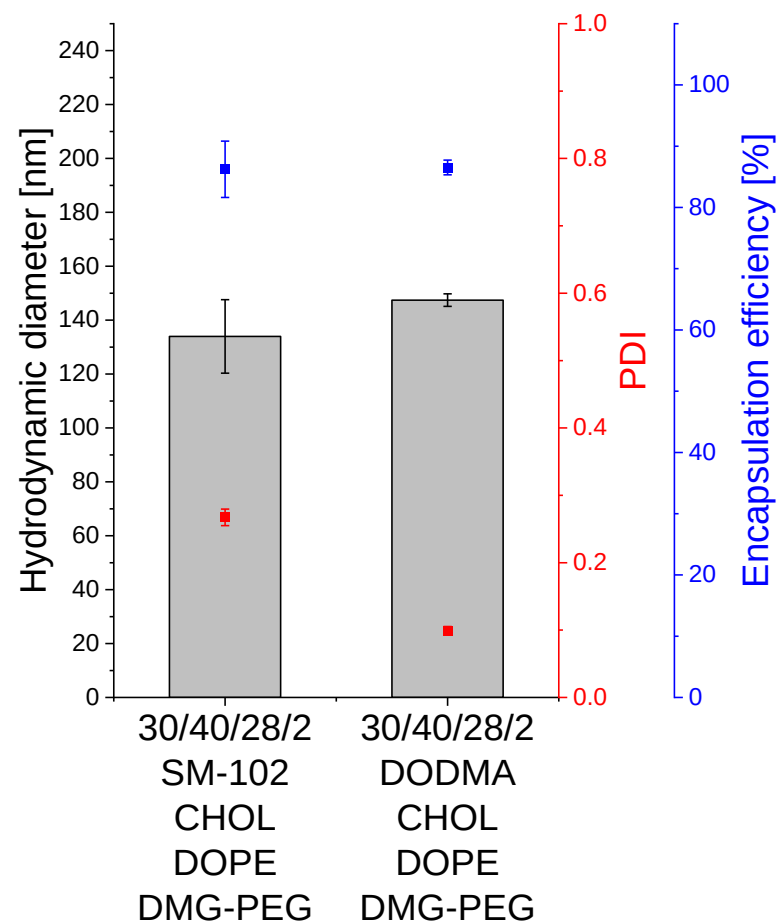


Preparing mRNA Loaded LNPs



Preparing mRNA Loaded LNPs

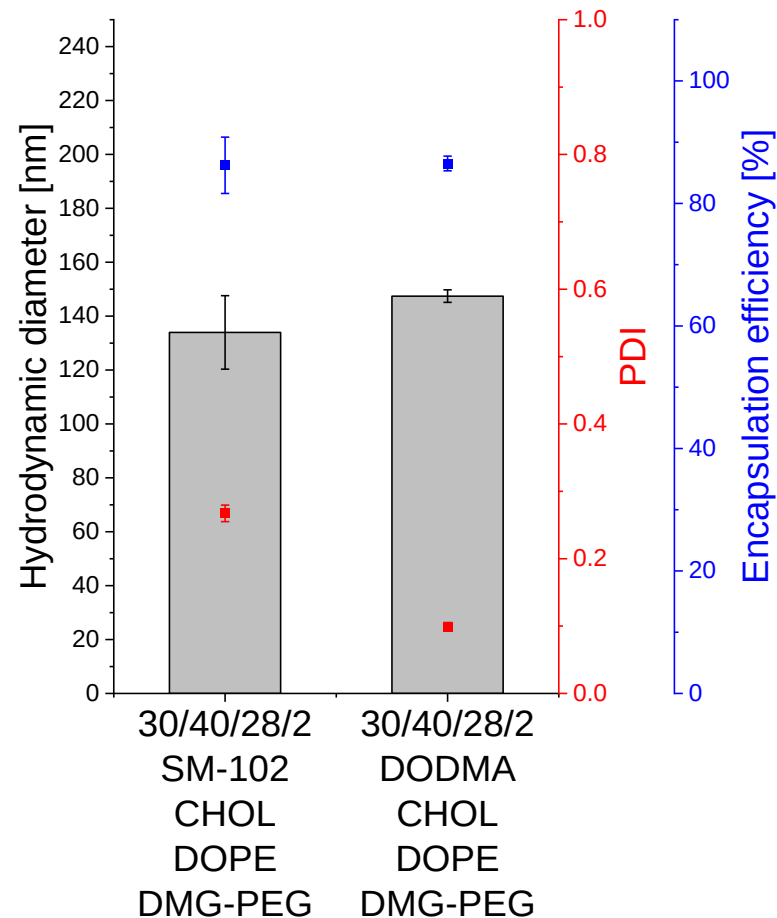
Data shown as
mean $\pm$ SD (n=3)



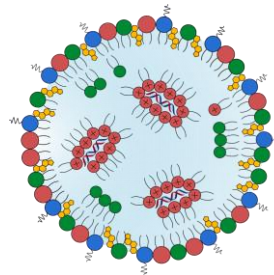
LNP Characteristics

- mRNA payload
- Lipid mass 1 mg

Preparing mRNA Loaded LNPs

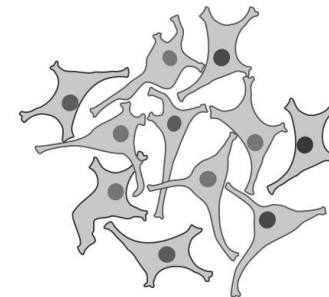
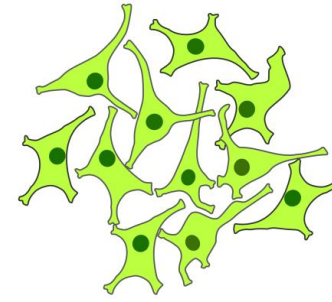


HeLa cells
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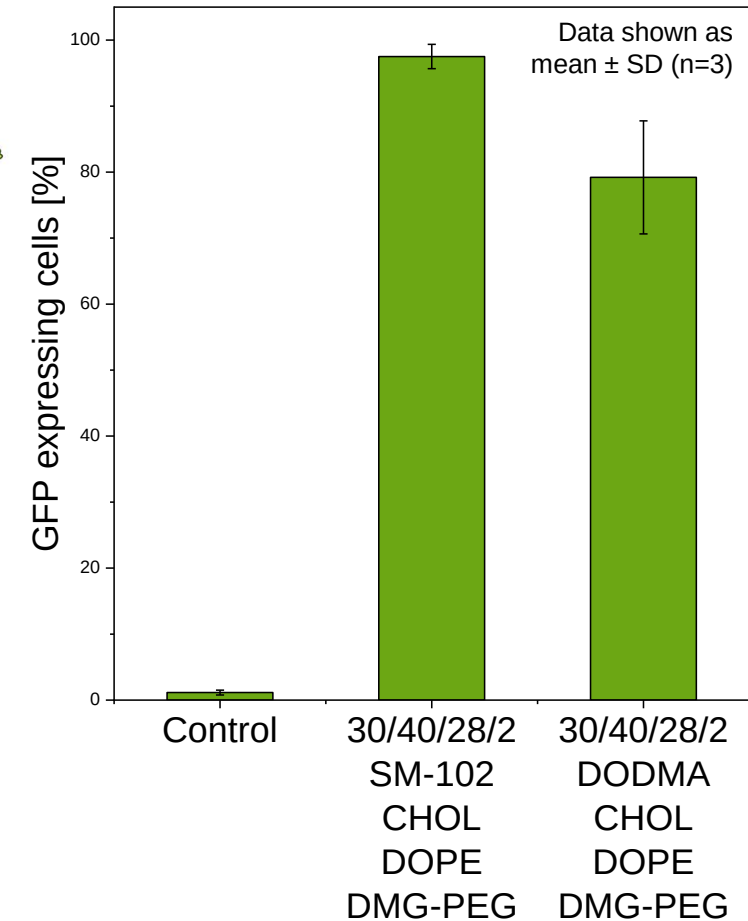
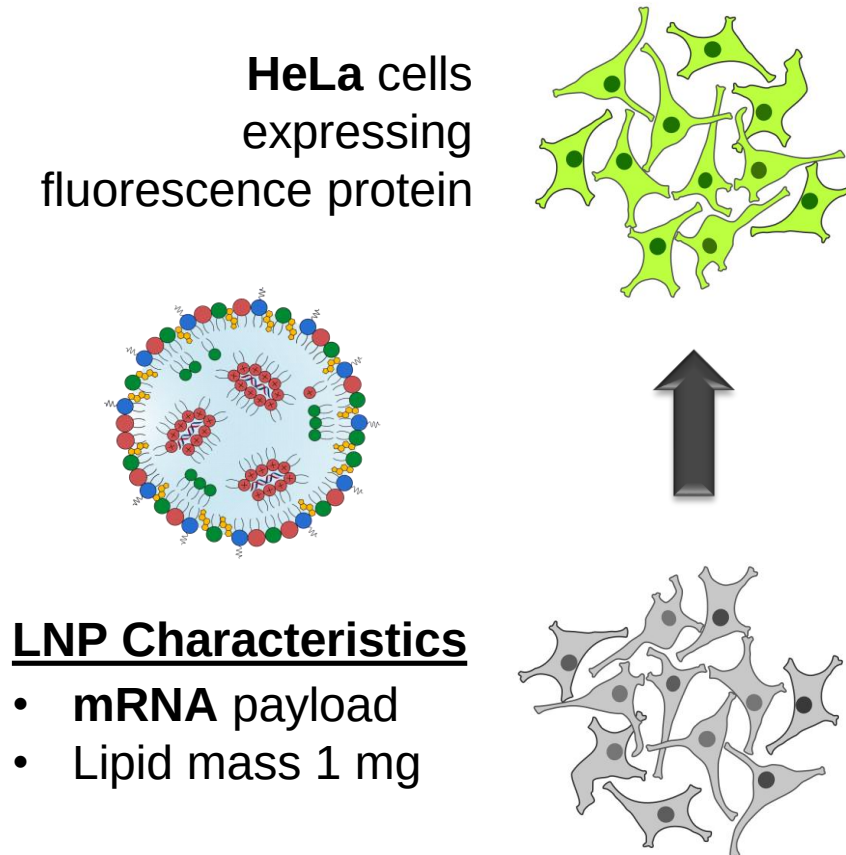
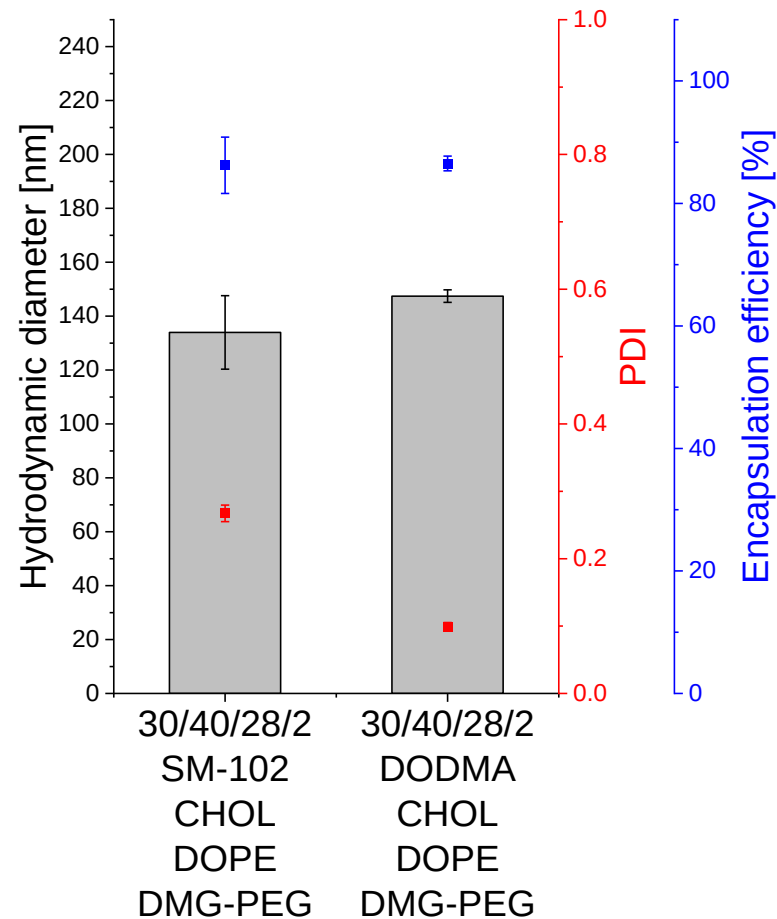
LNP Characteristics

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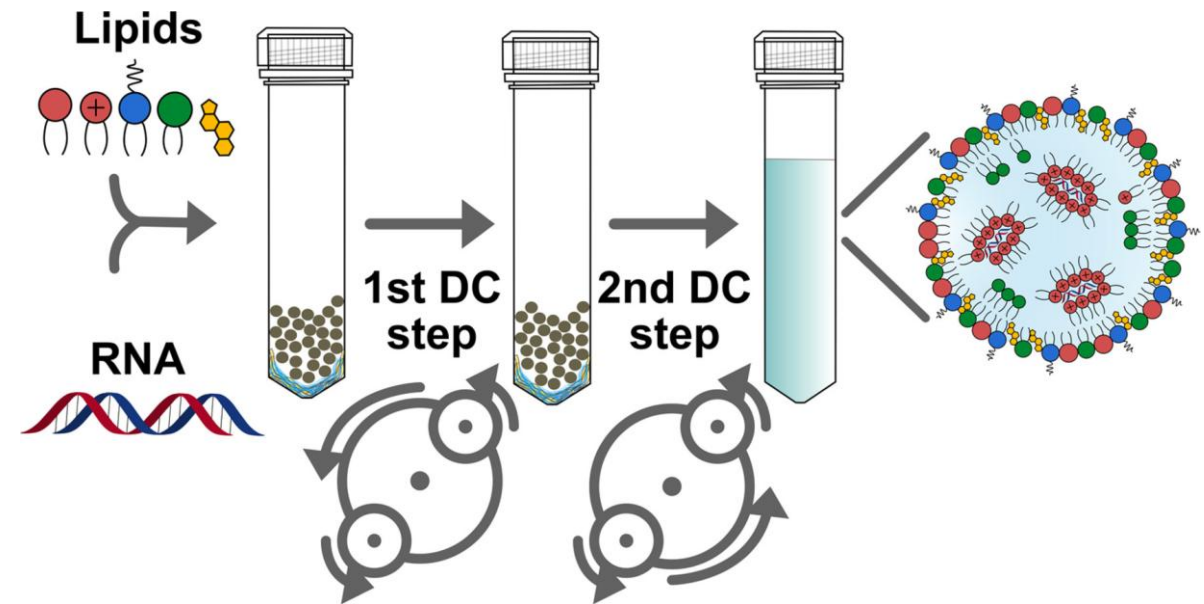
Data shown as
mean $\pm$ SD (n=3)

Preparing mRNA Loaded LNPs



Conclusion

- **High-throughput screening:** Enables efficient early-stage testing of RNA-loaded LNPs.
- **Miniaturized work scale:** lipid mass down to 1 mg.
- **Patient individual bedside preparation:** A simple, cost-effective alternative to e.g. microfluidics for LNP production.



Acknowledgement



universität freiburg

Prof Regine Süss



Prof Ulrich Massing



 **Lipoid**

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