# Design spaces of Lipid Nanoparticles for effective mRNA therapy

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Lipid Nanoparticles (LNP) are versatile carriers for biomolecules, with a promising focus on the delivery of mRNA for treating and preventing infections.[1, 2] Achieving efficient, safe and tissue-dependent LNP delivery involves the incorporation of specific lipids or the precise adjustment of the LNP's physicochemical properties, such as size, morphology and zeta potential.[3]

The development of novel targeting LNP platforms introduces formulation challenges, as variations in the lipid composition or microfluidic production parameters can significantly impact critical quality attributes (CQA) such as encapsulation efficiency, size or polydispersity index. In response to this challenge, we implemented a Design of Experiment (DoE) to efficiently screen critical process parameters (CPP) during the LNP microfluidic production of two model formulations: Comirnaty™ and Spikevax™. The obtained LNP formulations were investigated for *in vitro* cell transfection efficiency across various cell lines, facilitating the identification of optimal design space. The resulting correlation of CPPs and CQAs of the LNPs together with the cell transfection knowledge can be used to produce tuneable LNPs with predictable properties, while ensure reproducibility and consistent quality during the production process.

Further work will use this acquired knowledge as a foundation for the development of an automated and self-learning Microfluidic system, empowering high-throughput screening of novel lipids, enhancing the versatility and adaptability of the LNP systems.

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(1) Schoenmaker, L. *et al.* mRNA-lipid nanoparticle COVID-19 vaccines: Structure and stability. *International Journal of Pharmaceutics* **2021**, *601*, 120586. DOI: 10.1016/j.ijpharm.2021.120586.
(2) Zhang, Y. *et al.* Opportunities and Challenges for mRNA Delivery Nanoplatforms. *The Journal of Physical Chemistry Letters* **2022**, *13* (5), 1314-1322. DOI: 10.1021/acs.jpclett.1c03898.
(3) Kranz, L. M. *et al.* Systemic RNA delivery to dendritic cells exploits antiviral defence for cancer immunotherapy. *Nature* **2016**, *534* (7607), 396-401. DOI: 10.1038/nature18300.