Assessing the size and particle concentration of LNP using Videodrop: a new metric for LNP development

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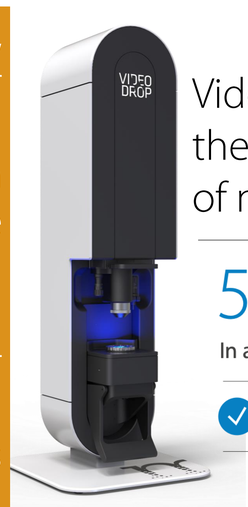
Neovacs develops mRNA-based therapies targeting chronic inflammatory diseases, such as allergies. Accurate characterization of lipid nanoparticles (LNPs) is key for ensuring stability and efficacy during manufacturing and storage.
Videodrop, developed by Myriade, is a label-free technology based on Interferometric Light Microscopy (ILM), allowing real-time measurement of particle size and concentration at the single-particle level.
In this collaborative study, Videodrop was used to evaluate:
- **Process development**: comparing filtration vs. tangential flow filtration (TFF) for impact on LNP intgegrity.
- **Stress and stability conditions**: thermal and mechanical stress to study LNPs degradation.

These results support Videodrop as a fast, sensitive, and user-friendly tool for LNP development and process monitoring.



Videodrop measures the **size and concentration** of nanoparticles.

Virus | EVs | LNP

5-10µL In a single drop | 80-500nm Size range | 10^8-10^{10} part/mL Concentration range | UP TO 10µm Visualization

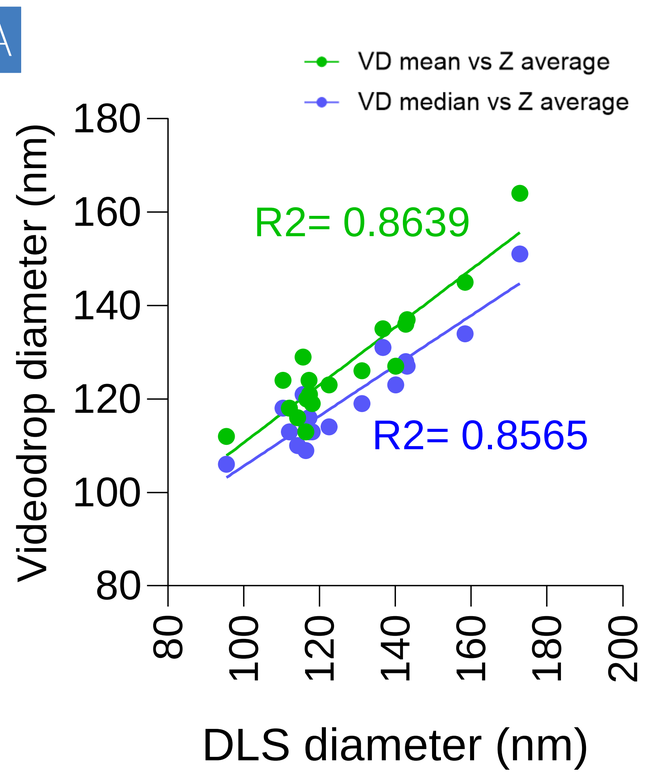
21CFR part 11 compliant | No labelling & non denaturant | Complex & viscous sample

Material & Methods

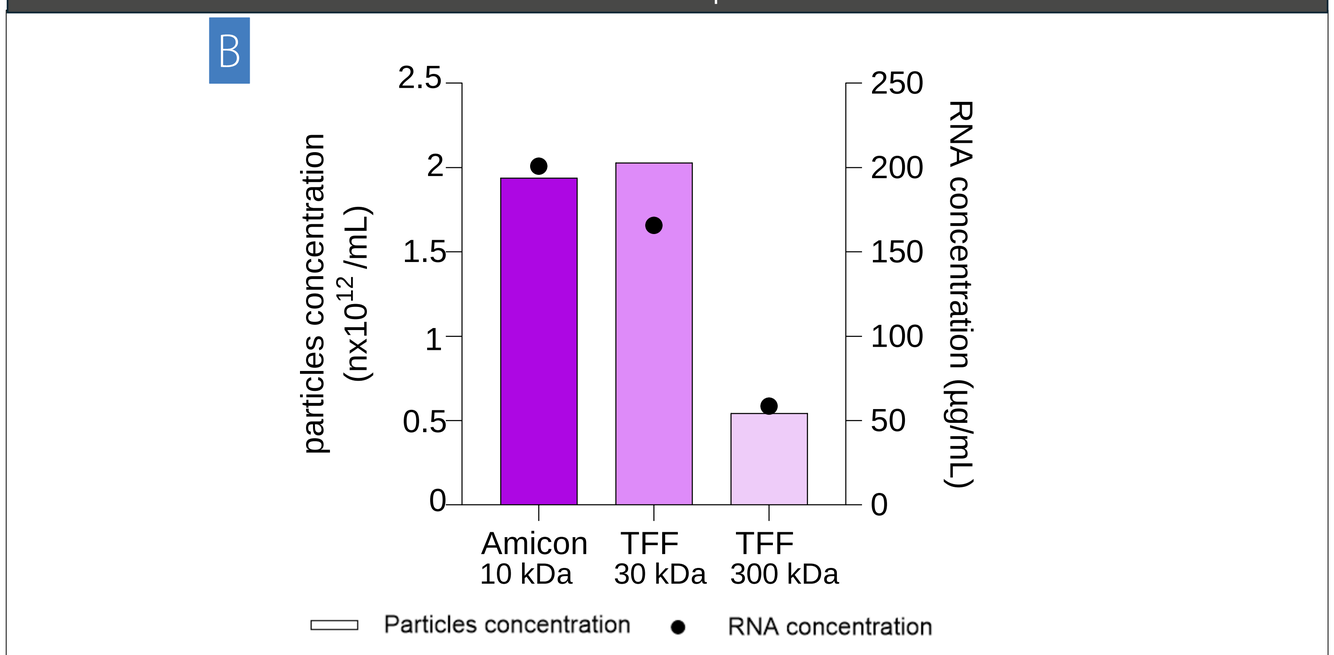
First, we compared LNPs hydrodynamic diameters measured by Videodrop and DLS (Zetasizer µV, Malvern Panalytical, UK) to ensure the accuracy of Videodrop measurements (n=18 LNPs).
Then, we evaluated the impact of purification process, stress (mechanical, Freezing (-80°C)/Thawing and temperature (37°C)) on size and particles concentration of LNPs using Videodrop.
LNPs were produced by Neovacs, from different mRNA sequences, LNP compositions and formulation buffers. RNA concentration were determined by fluorescence Ribogreen assay.

Results

In figure A, we compared Videodrop mean and median diameters versus DLS Z average.
A positive correlation is shown (spearman correlation, with $R^2 > 0.8$). These results demonstrate that Videodrop may be used to easily and quickly control LNP size throughout the manufacturing process.

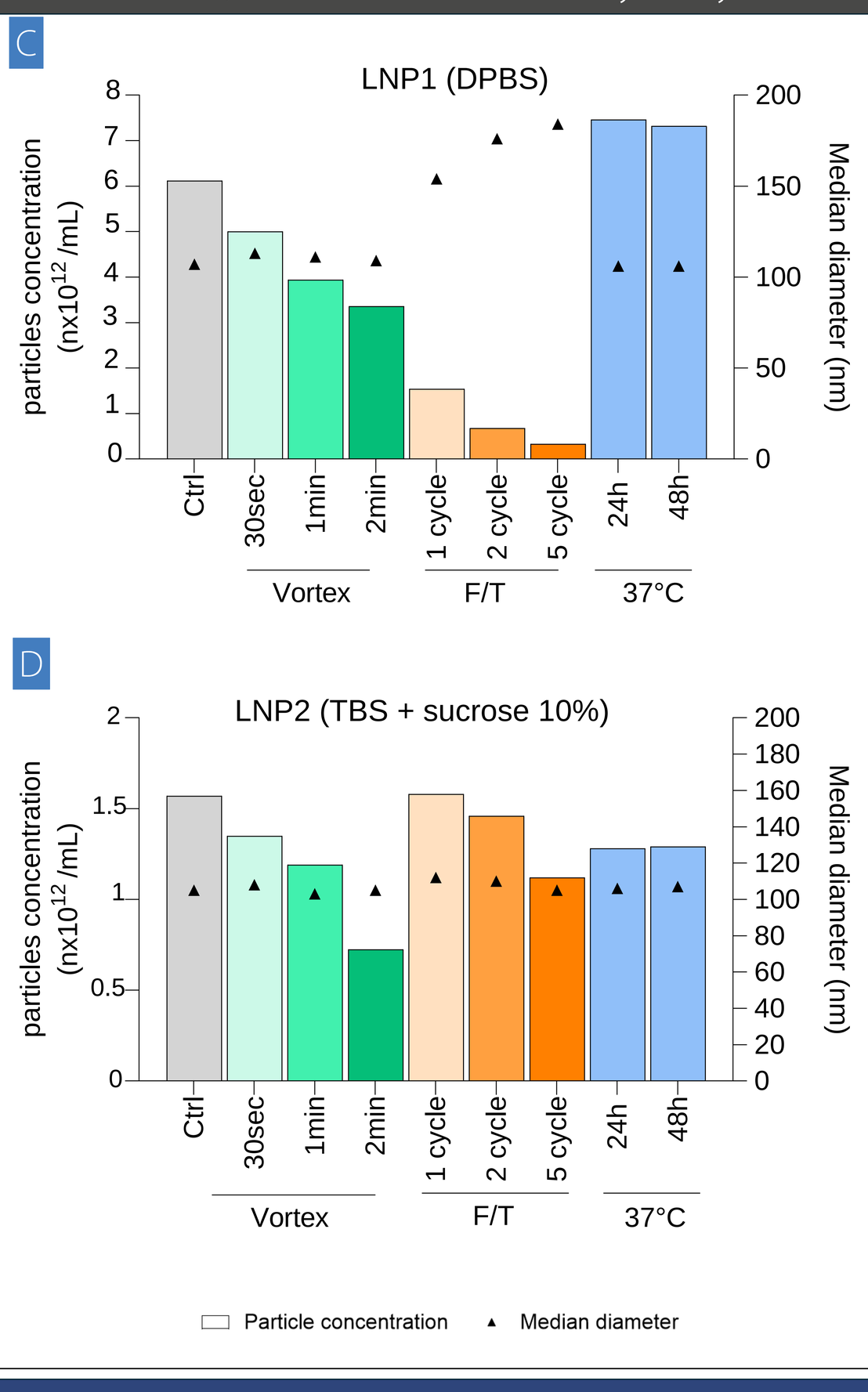


Purification process



In figure B ,Videodrop is used to evaluate the impact of different purification systems on LNP yield. This figure shows that Ultrafiltration and TFF with a 30 kDa cut-off are equivalents, whereas TFF with a 300kDa cut-off results in a loss of particles.
By comparing total RNA concentration measured using the Ribogreen assay and nanoparticle concentration, we conclude that TFF with a 30 kDa cut-off is more suitable for LNPs purification without loss of nanoparticles through the membranes.

Stress, formulation and stability study



In figures C and D, we evaluated the impact of vortexing and temperature stress (Freeze/Thaw and storage at 37 °C) on particles size and concentration of distinct LNPs formulated in DPBS or TBS+sucrose 10 %.
Vortexing reduces the number of LNP particles whatever the formulation buffer depending on the duration, but do not affect LNPs size.
F/T cycles results in a significant drop in the number of particles with an increase of LNPs size in the absence of sucrose (Fig C). The addition of sucrose induce cryoprotective effect of LNP (Fig D), preventing aggregation/degradation phenomenom under these conditions.
Finally, storage at 37 °C up to 48 hours has no impact on the particle concentration and size of the two LNPs.

Particle concentration becomes an important quality criteria to include in the **LNP quality analytical strategy**.
Videodrop is an ideal companion tool for LNP process and formulation development.