

# NanOZ LNP-siRNA Results

## Storage and shipping conditions.

Storage: -80°C upon reception and for long-term use.

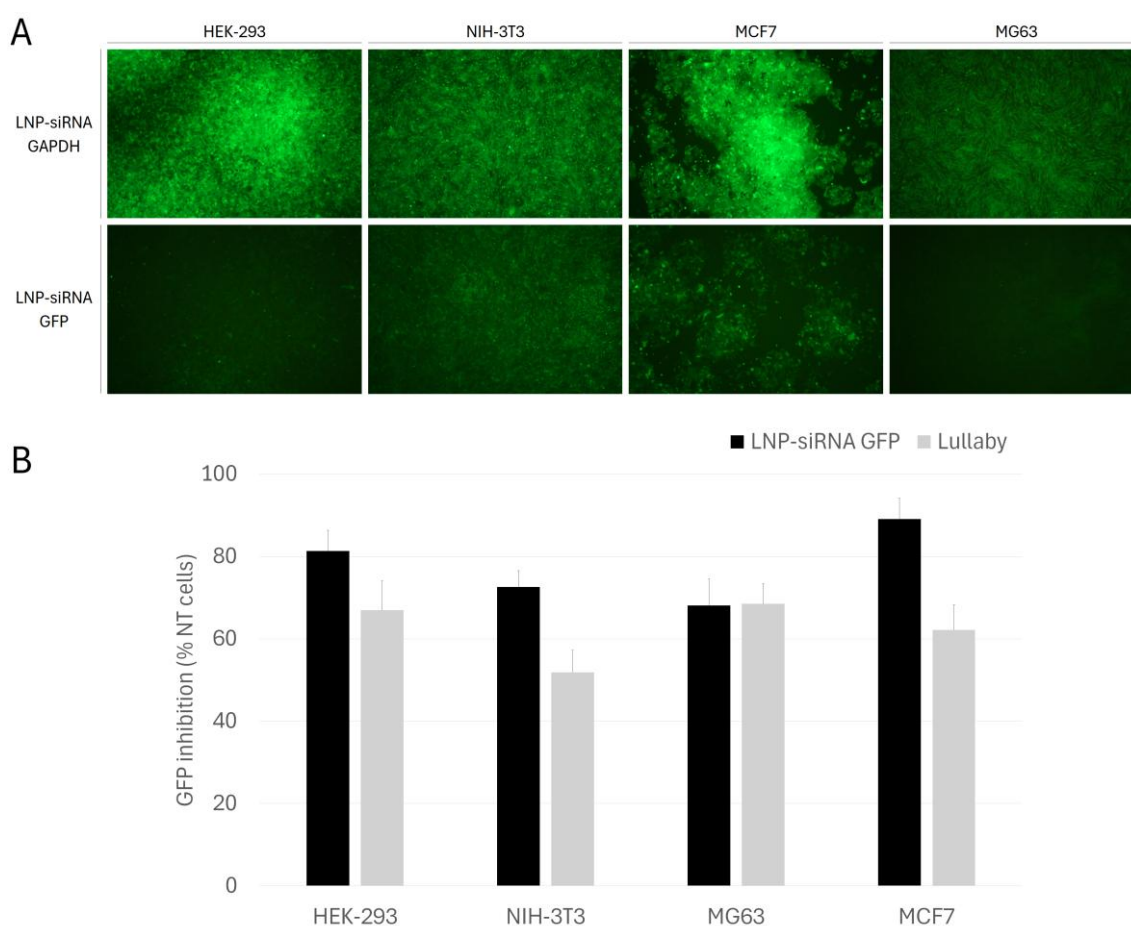
Stability: 6 months.

Shipping condition: Dry Ice.

## NanOZ LNP-siRNA GFP for gene silencing

### NanOZ LNP-siRNA allows efficient siRNA mediated gene silencing in cell lines.

LNP encapsulating 400 nM siRNA were formulated through pressure-driven controlled microfluidic systems. 5 nM LNP-siRNA GFP and LNP-siRNA GAPDH were added to cell lines expressing GFP and inhibition of fluorescence was evaluated after 72H.



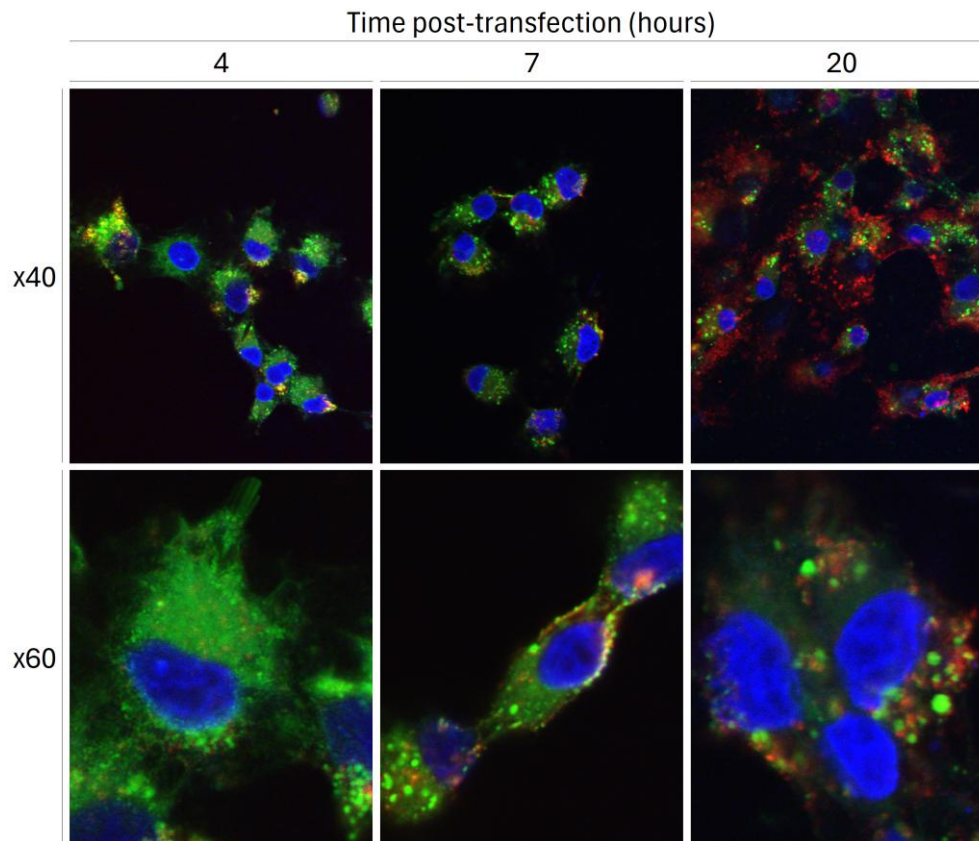
**Figure 1: Gene silencing in various cell lines:** GFP stably expressing cells were transfected with 5 nM siRNA against GFP encapsulated in LNP (LNP-siRNA GFP) or complexed with Lullaby (Lullaby) or 5 nM siRNA against GAPDH encapsulated in LNP (LNP-siRNA GAPDH). 72H after transfection, inhibition of fluorescence intensity was observed under fluorescence microscopy (A) and % of inhibition was calculated against Non Treated cells (NT cells) by flow cytometry (B).

Results demonstrate the capacity of LNP-siRNA GFP to efficiently silence gene expression in cell lines.

# NanOZ LNP-siRNA Delivery

## NanOZ LNP efficiently deliver siRNA.

LNP were prepared as described previously by microfluidic mixing of an ethanolic lipid phase using DiO labelled lipid with an acidic aqueous phase containing Cy5 siRNA to follow delivery in cell line following transfection. The resulting dual-color system enables real-time visualization of both the carrier and its cargo, as shown in the figure below.



**Figure 2: NanOZ LNP mediates efficient siRNA delivery:** Cy5 non targeting siRNA (Red) was encapsulated in Dio NanOZ LNP and added to HEK-293 cell line. Delivery was followed for 20 hours under confocal microscopy at two magnification (x40 and x60), DAPI counter staining was used to localize cell nuclei.

Results demonstrate the capacity of LNP-siRNA to efficiently deliver its cargo in the cytoplasm. 20H after transfection we can see that the siRNA is almost fully released in the cytoplasm.