Lipid Nanoparticle Formulation Characterization: Bridging the Gap from R&D to GMP

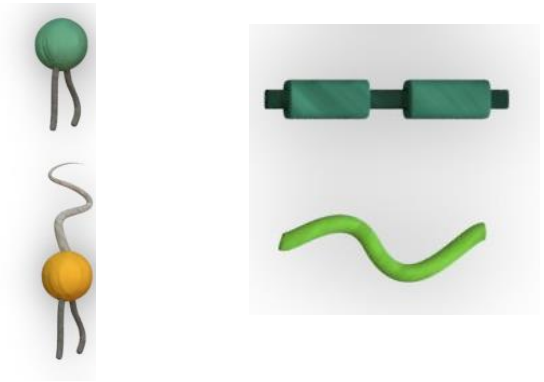
SERGIO ESTEBAN PÉREZ

CONTROLLED RELEASE SOCIETY
CRS 2024 Annual Meeting AND Exposition
JULY 8-12, 2024 • BOLOGNA, ITALY

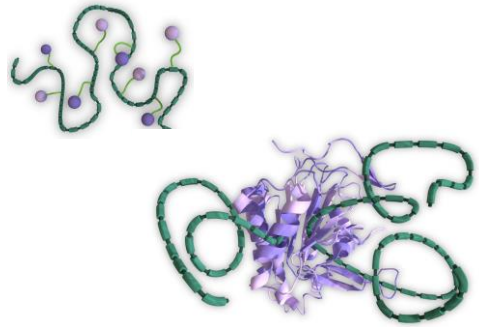
INTEGRATING
Delivery Science
ACROSS DISCIPLINES

Enabling next-generation Non-Viral Gene Therapy through custom design and end-to-end services in manufacturing of polymer and lipid-based therapeutics.

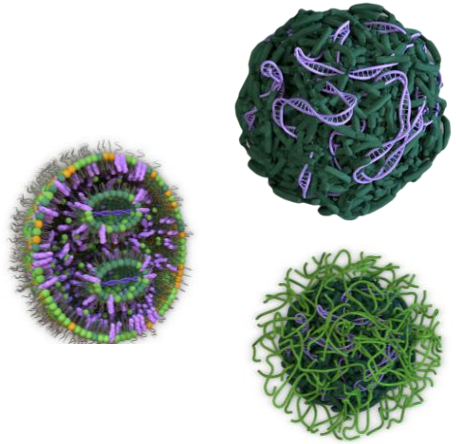
Our product categories include:



Polymer & Lipid Excipients



Chemical and Biological Conjugation



Lipid and Polymer Nanoparticle Formulation

Aseptic Fill & Finish

With our integrated partnership style, we support small and large clients and become a strategic partner to solve challenges in development and accelerate your speed to the clinic.

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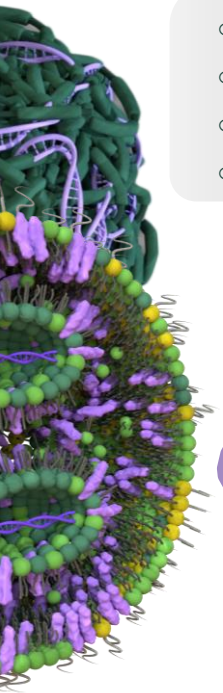
We smoothly navigate your therapeutic product throughout the clinical stages

RA Based & Phase Appropriate CMC Development for Novel Excipients & Nanoparticle Drug Products



Tech Transfer
30-60 Days

- Scientific Review
- Tech Transfer
- GAP assessment
- Comparability studies

Preclinical
80-150 Days

- Syn. Route Development
- Formulation Developmentn
- Definition of TPP & key CQAs
- Method Development
- Pilot Scale-up (to Kg)
- Non-Clinical Material Supply

IND Enabling
120 Days

- Process characterization
- Preliminary control strategy
- Method Qualification
- Impurity profiling
- Formal ICH Stability
- GMP Clinical Trial Supply
- Regulatory CMC package

Phase I-II
120-200 Days

- Inventory management
- Continued process Characterization
- Updated Control Strategy
- GMP CTM supply(1-6Kg batch)
- Continuous Regulatory Support

Pivotal Phase, Commercial supply
300-500 Days

- Process Validation
- Method Validation
- Final Control Strategy
- Formal ICH Stability
- BLA filling support
- GMP CTM supply
- Commercial Supply

8 INDs submitted

Supporting **6** Clinical Trials

2 Commercial products supplied

5 weeks Average batch release time

+130 GMP Batches

100% Batch Compliant Rate

+150 Methods Validated ICHQ2

+200 Non Compendial Methods developed

100% Audit Compliance

>93% Project OTIF rate

30% PhDs

PMO Under PMP® program

56 cNPS

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Collaborating with pharma and biotech companies worldwide

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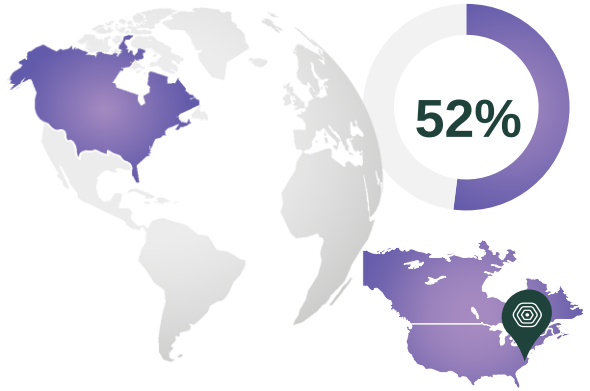
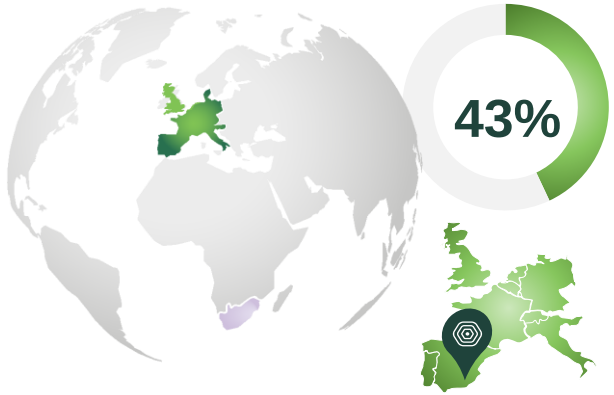
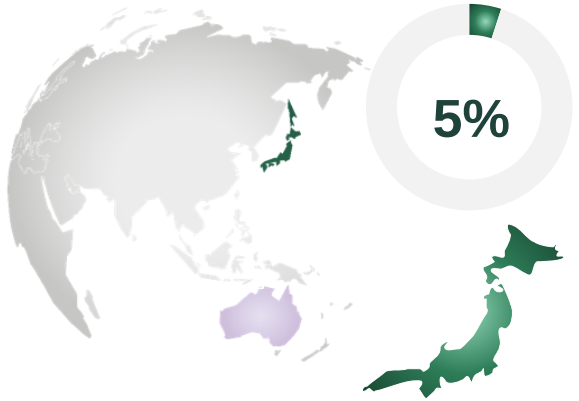
North America



Europe



Japan

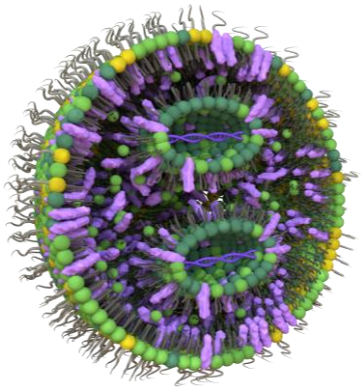


Product Type %	
PNP	51%
LNP	29%
Polymer Conjugate	17%
Polymer Drug	3%

Payload %	
Nucleic Acid	35%
Small molecule	28%
Other	25%
Biologic	12%

Candidate Stage %	
Discovery-preclinical	74%
Clinical Stage	12%
Pivotal Stage	8%
Commercial	6%

Beyond PEGylated lipids: Non-immunogenic shielding lipids in vaccines



Ionizable Lipid *(neutral/protonated)*
Aids in the encapsulation of nucleic acids through electrostatic interactions

Cholesterol
Decreases permeability of the LNP and enhances its stability

Nucleic Acid *(e.g. mRNA)*
Encodes protein of interest

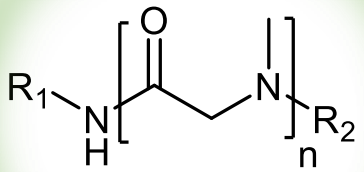
'Helper' Lipid
Improves LNP stability and fusogenicity

PEG-Lipid
Prevents non-specific protein absorption, particle aggregation and controls LNP size

PEG Alternatives
PSar, Biocompatible, endogenous building blocks, overcome PEG immunogenicity

The key challenge is to obtain a scalable and process developed LNPs formulation manufacture

Psar-based Lipid



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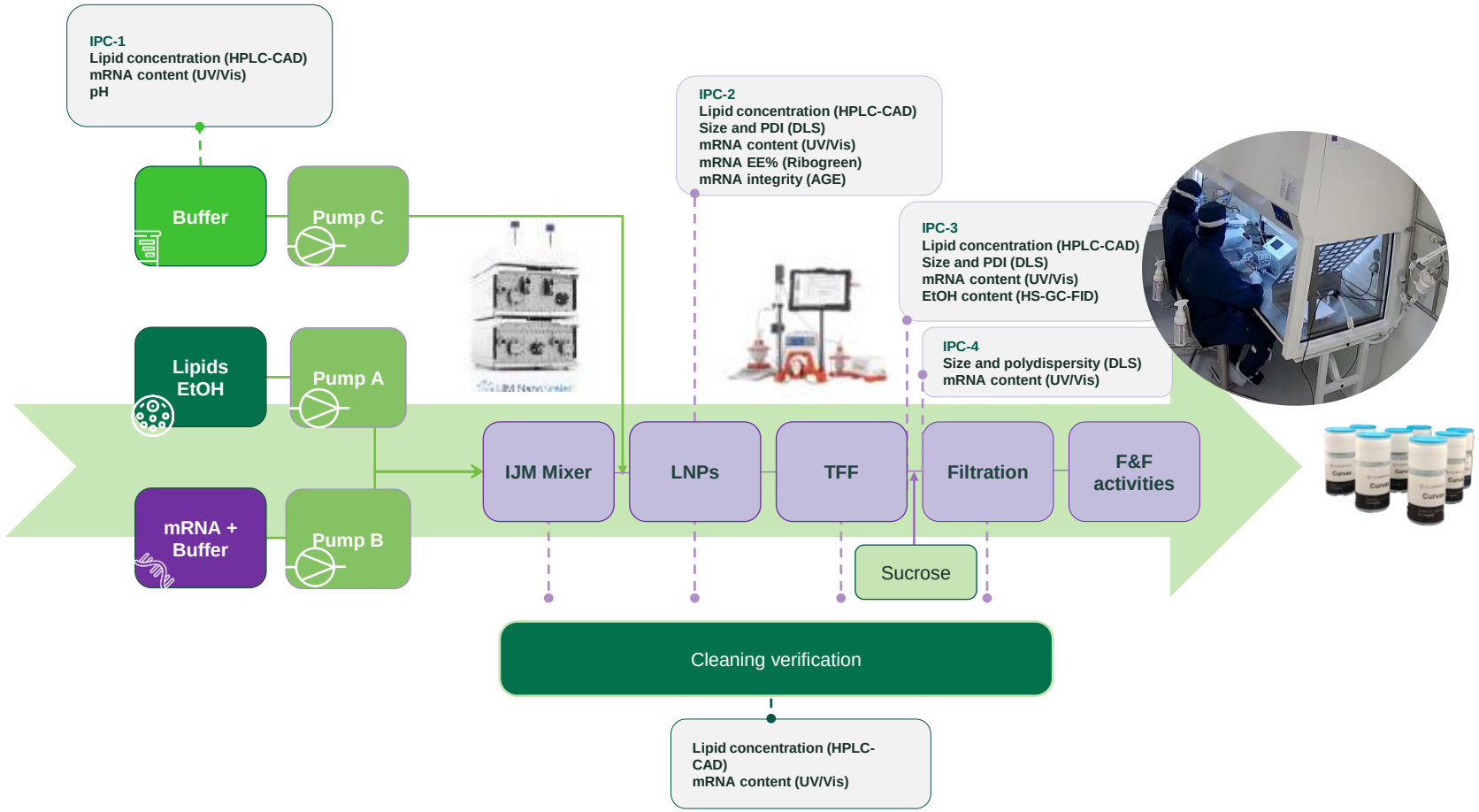
Integrating Process Development and Scale-Up



LIPID COMPONENTS	SM-102	DSPC	Cholesterol	DMG-PEG (2000)
mRNA COMPONENT	CatPure™ Firefly Luciferase mRNA			
BUFFER COMPONENT	Tromethamol hydrochloride	Acetic acid	Sodium acetate	Water for injection
CRYOPRESERVANT	Sucrose			
SCALE & API CONCENTRATION	1 L formulation 0.2mg/ml of mRNA (Luc)			
VIAL FORMAT	10R vials 6.3 mL filling volume (160 vials)			

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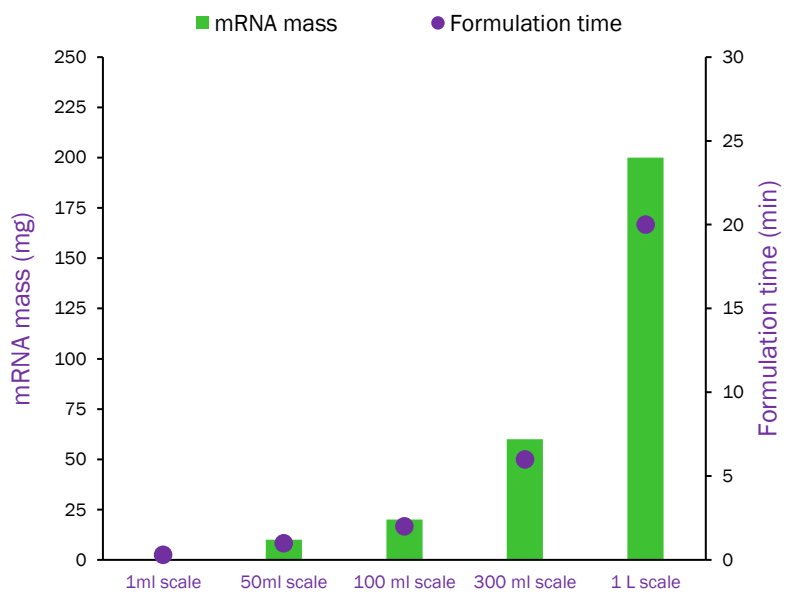
LNPs Formulation Process Development → Scale up process

Particle Size Distribution

Process Parameters



During the whole process development step the Particle size distribution remained controlled, allowing a scalation of the mRNA amount employed and optimizing the formulation time. The amount of mRNA and its encapsulation efficiency was monitored as well and remained constant

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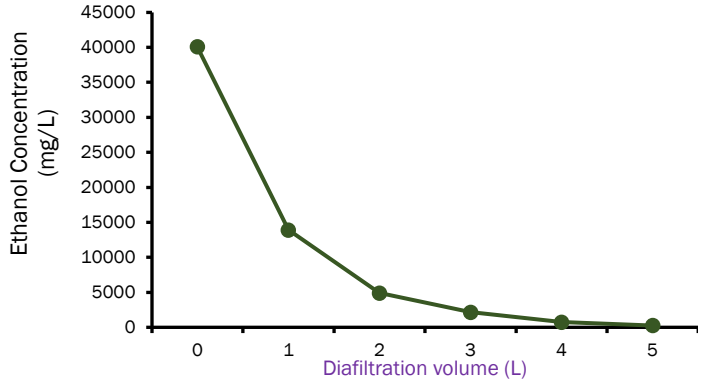
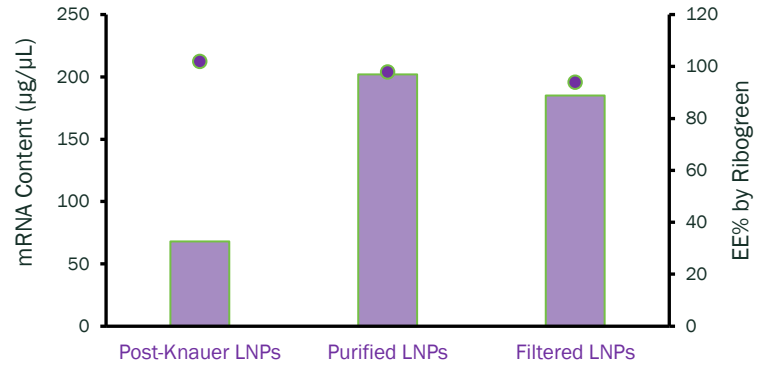
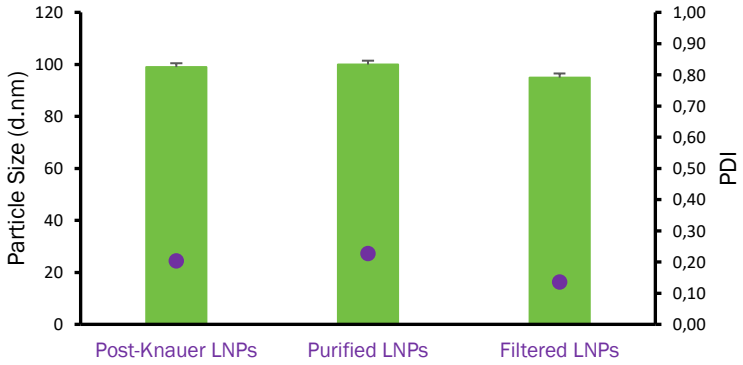
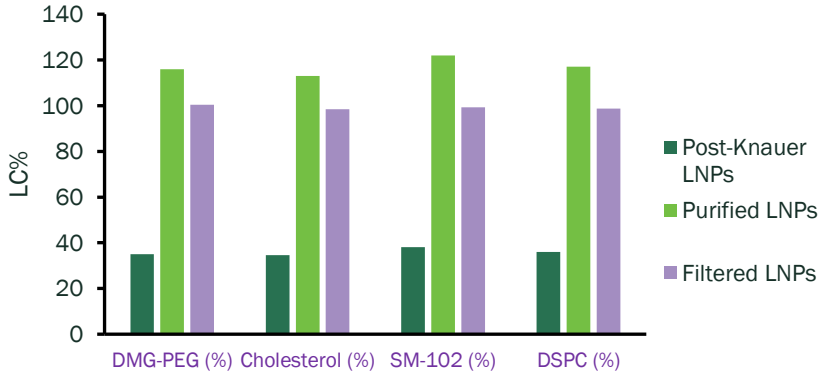
LNPs Formulation GMP Batch Preparation Critical Quality Parameters monitorization









The developed LNPs maintained their CQAs throughout the process and did not suffer alteration during the concentration and purification process

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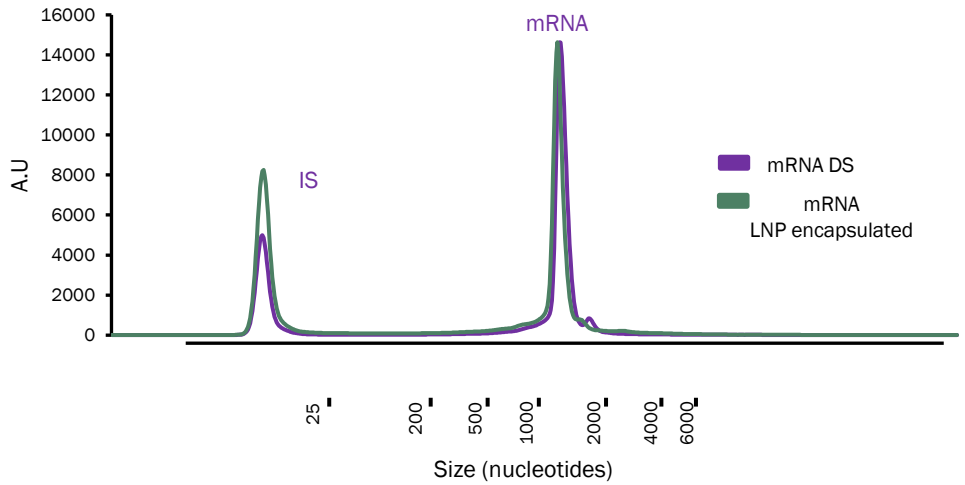
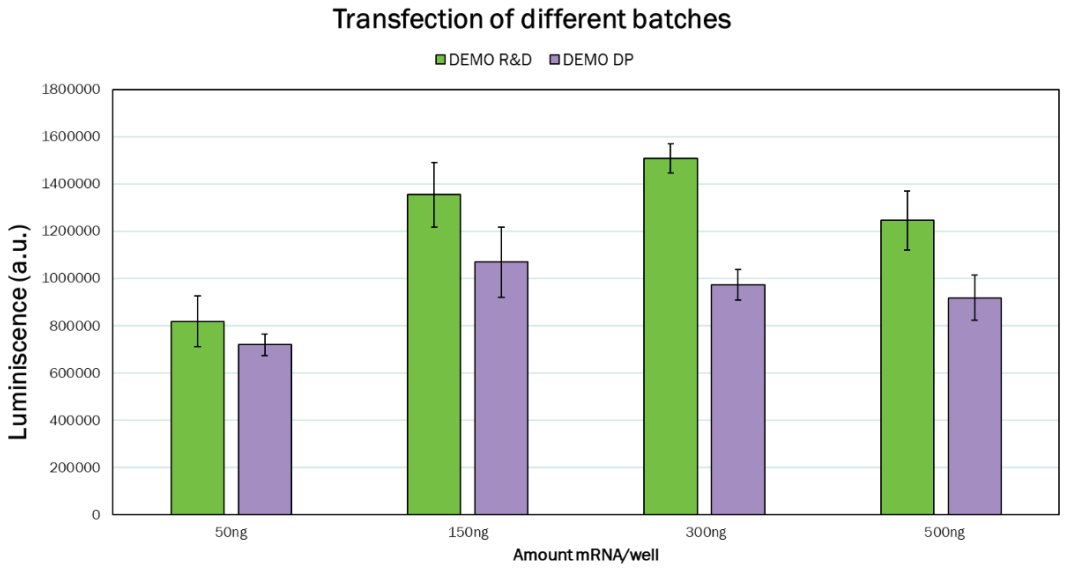
LNPs Formulation GMP compared with R&D Batch *In Vitro* efficiency



The mRNA integrity was retained after the encapsulation process



Both formulations presented the ability to transfect *in vitro*

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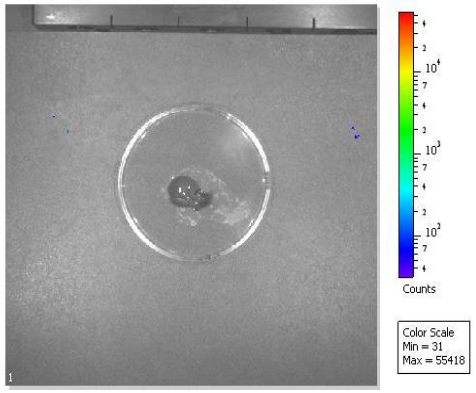
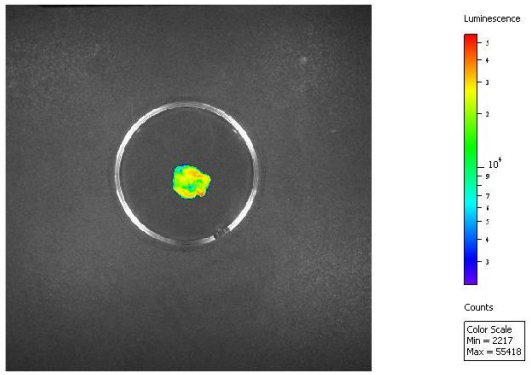
LNPs Formulation GMP compared with R&D Batch *In Vivo* efficiency

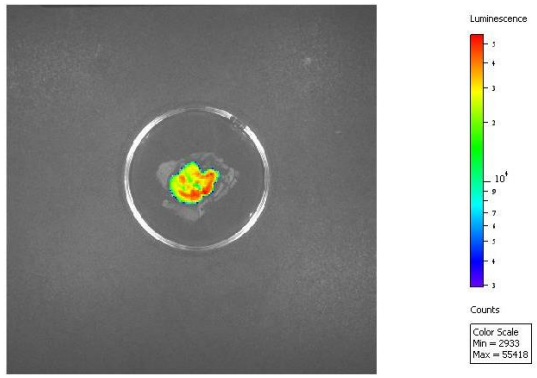
Control (PBS)

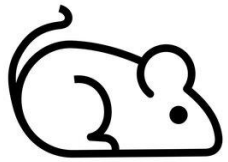


GMP LNPs

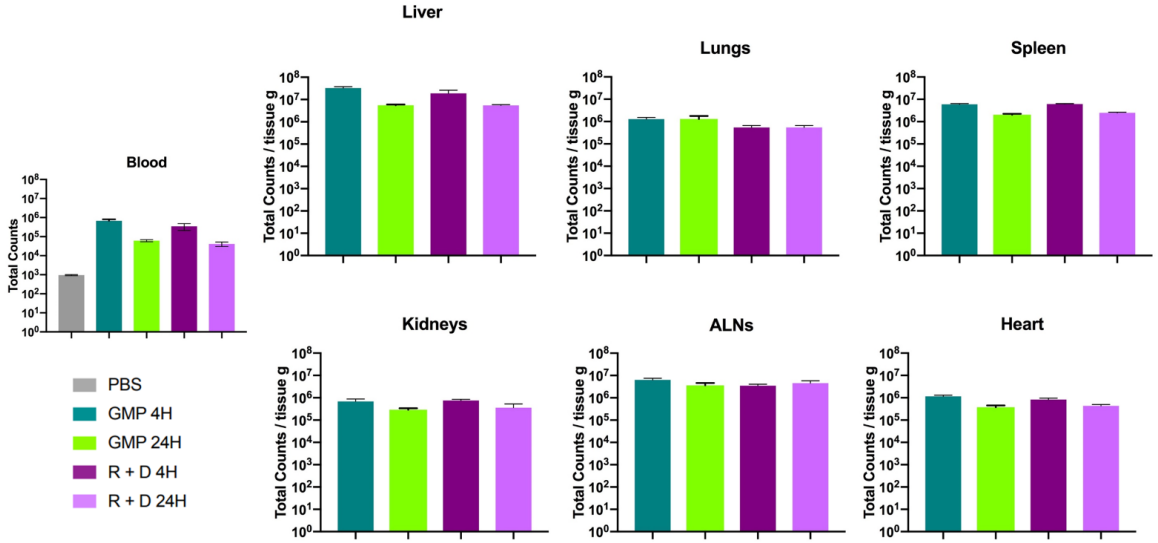


R&D LNPs





i.v.



Equivalent 5mL R&D vs 1L GMP batch *In Vivo* Expression

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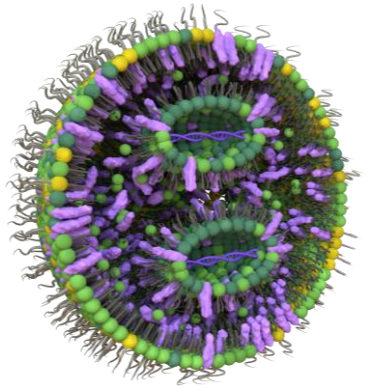
LNPs Formulation GMP final product Release



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Curapath
Parque Tecnológico
Av Benjamin Franklin 19, 46980 Paterna, Valencia (Spain)

Certificate of Analysis

Product Name: Curvax
Product Number: 1368
Storage temperature: -25°C to -15°C
Batch: 23-006
Manufacturing date: 1/Nov/2023
Analysis date: 30/Nov/2023

Parameter	Method	Limits	Results
Appearance	Visual inspection	White to off-white dispersion	White dispersion
Visible particles	USP <790>	Not detected	Not detected
pH	Ph.Eur. 2.2.3.	7.0-8.0	7.4
Osmolality	Ph.Eur. 2.2.35.	280-320	300
Extractable volume	USP <698>	Conforms	Conforms
Container closure integrity	USP <1207>	Conforms	Conforms
Size (nm)	DLS	90-110	95
Polydispersity index	DLS	< 0.20	0.14
DMG-PEG content (mg/mL)	HPLC-CAD	0.14-0.22	0.18 (100 %LC)
Cholesterol content (mg/mL)	HPLC-CAD	0.56-0.84	0.69 (98 %LC)
SM-102 content (mg/mL)	HPLC-CAD	1.34-2.01	1.66 (99 %LC)
DSPC content (mg/mL)	HPLC-CAD	0.30-0.45	0.37 (99 %LC)
Total RNA content (µg/µL)	UV-Assay	0.18-0.22	0.19 (95% LC)
RNA encapsulated (%) w/w	Fluorescence	> 70	94
Total unknown impurities (%) w/w	HPLC-CAD	< 10.0	2.4
Residual ethanol (mg/L)	GC-FID	< 5000	< 5000
In vitro expression	Assay	>750000 RLU	Conforms
Endotoxin content	Ph.Eur. 2.6.14.	<100 EU/ml	Conforms
Sterility	USP <71>	Conforms	Conforms

Date and Place: 30/NOV/2023, Paterna (Valencia)

Curapath warrants that at the time of the quality release or subsequent retest date this product conformed to the information contained in this publication. Purchaser must determine the suitability of the product for its particular use. Product can only be used for research purposes.

- The monitorization of the CQAs led to the obtention of a scalable LNPs manufacturing process
- The CQAs has remained unaltered during the whole process development stage
- The R&D and GMP formulation has provided similar performance *in vitro* and *in vivo*

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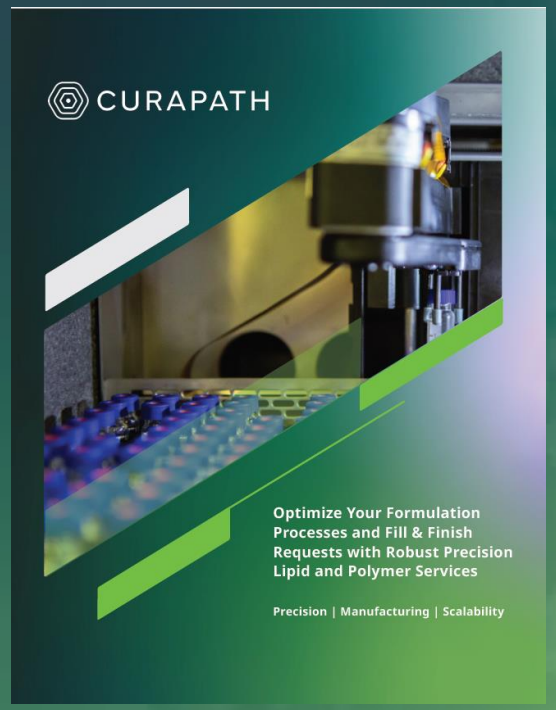
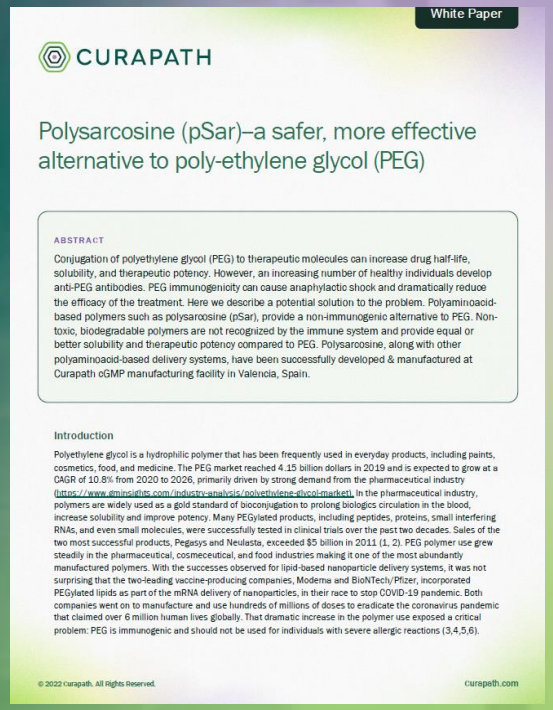
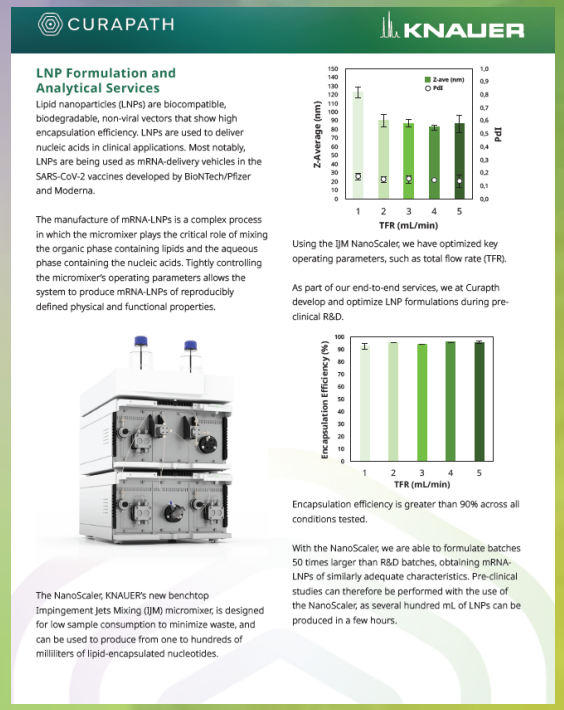
Brochures, Product Catalogue, Case Studies, Product Highlights, App Notes, Posters…..







… and more to come

Booth #61

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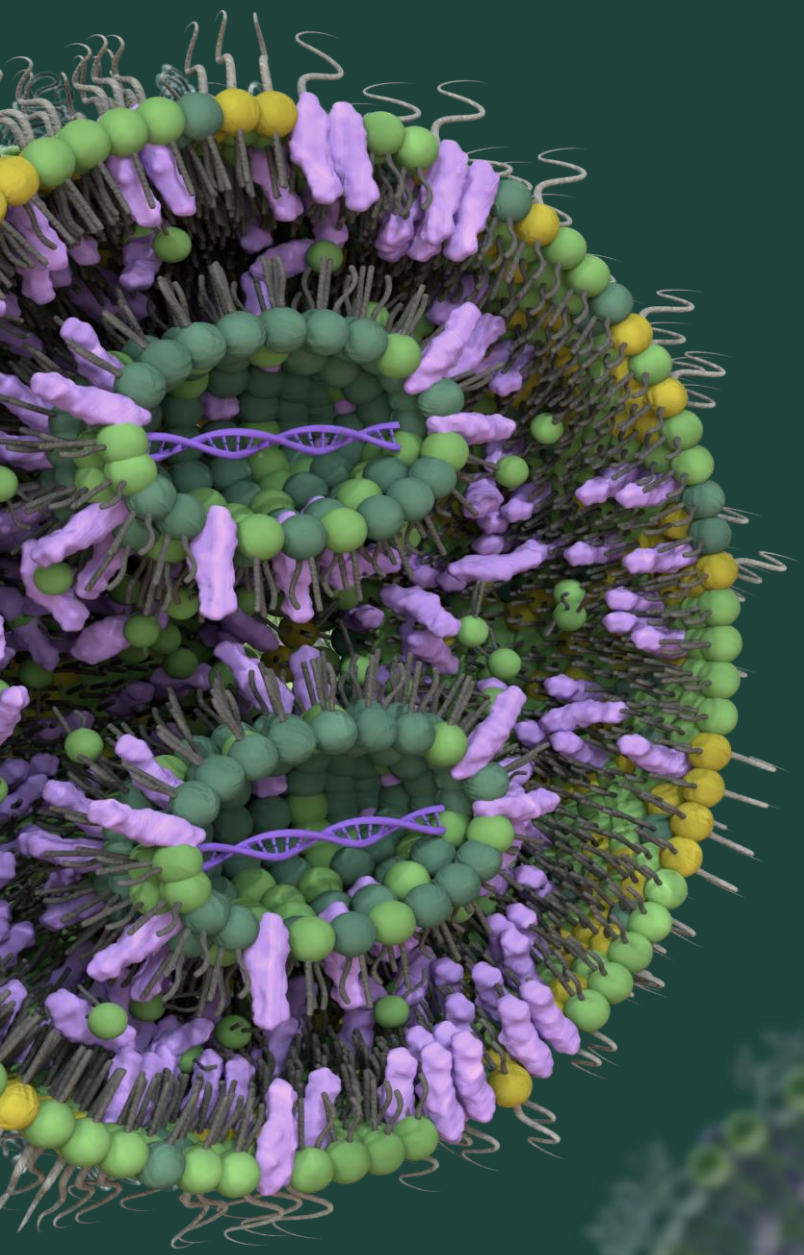

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Innovation & Development site

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