Enabling the next-generation non-viral gene therapy through Lipid-PEG alternatives based on bioinspired polymers

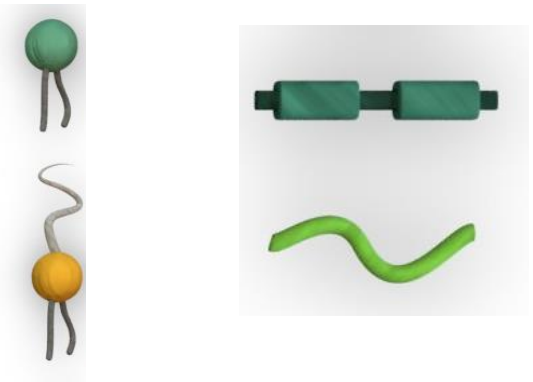
Aroa Duro Castaño, Curapath, Spain

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

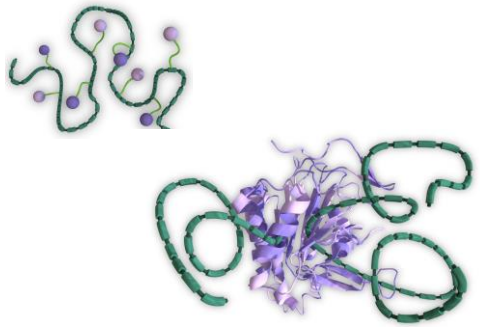
INTEGRATING
Delivery Science
ACROSS DISCIPLINES

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

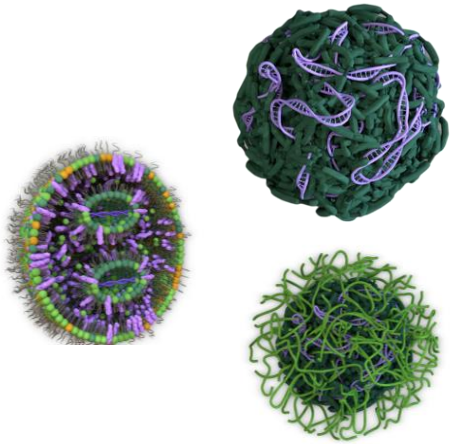
Our product categories include:



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.

CURAPATH

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

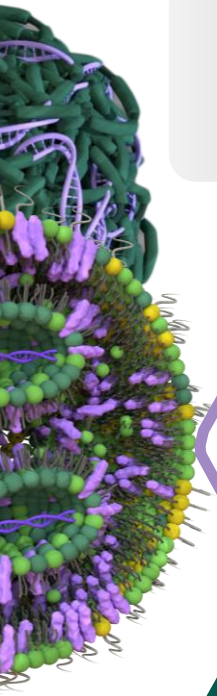
- Process characterization
- Preliminary control strategy
- Method Qualification
- Impurity profiling
- Formal ICH Stability
- GMP Clinical Trial Supply
- Regulatory CMC package

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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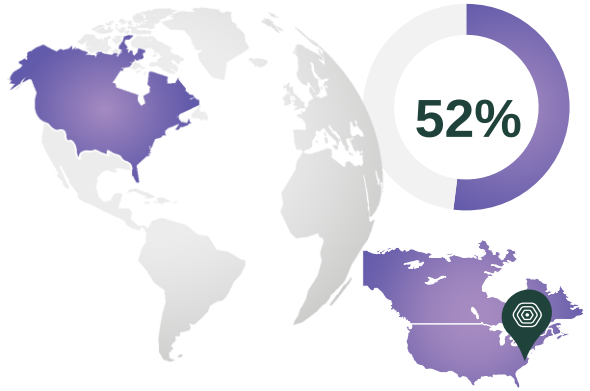
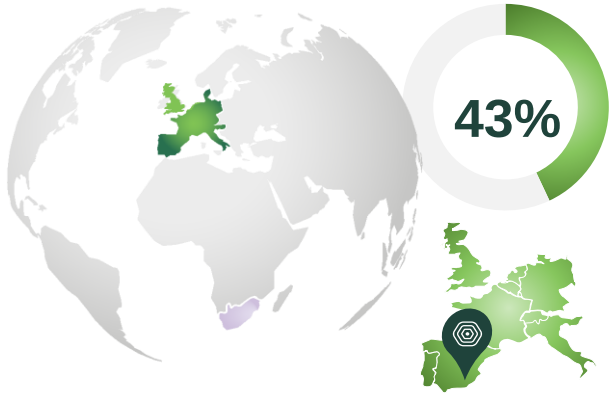
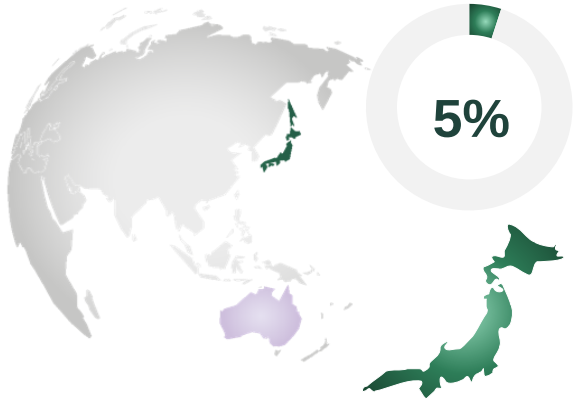
North America



Europe



Japan



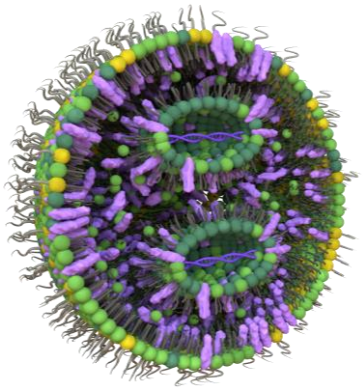
+50 Active Customer Accounts, world leading innovator Biotechs and Pharmaceutical companies

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

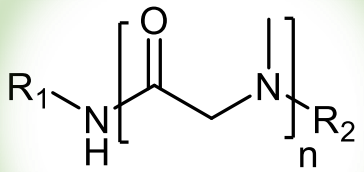
'Helper' Lipid
Improves LNP stability and fusogenicity

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 mimicking PEG physic-chemical properties while avoiding immunogenicity

Psar-based Lipid



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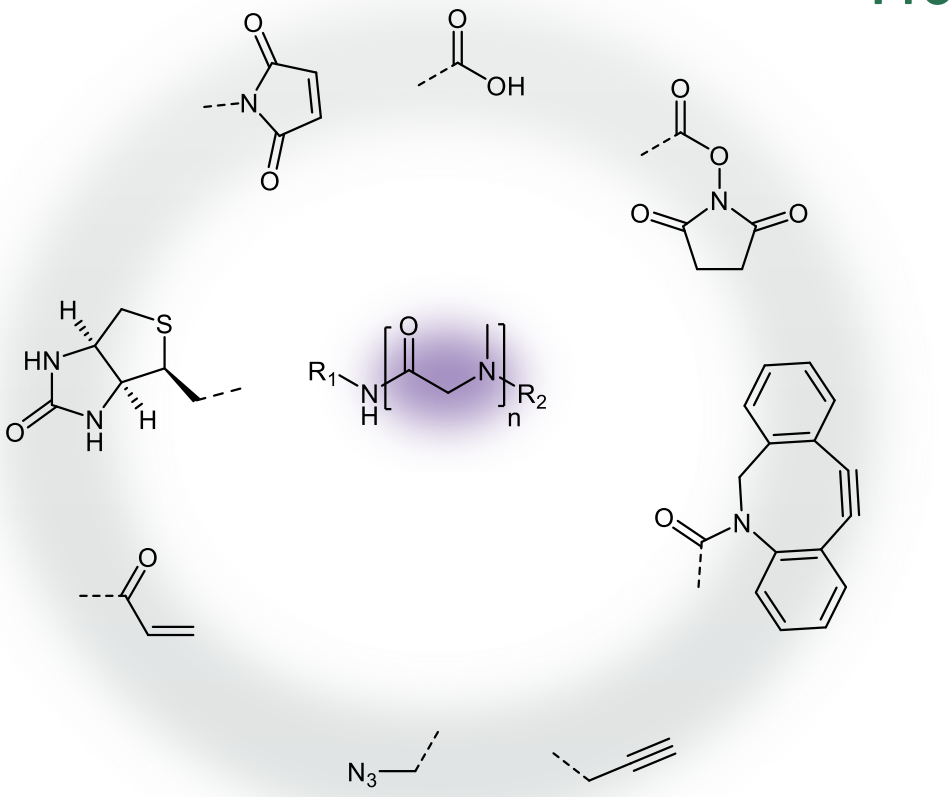
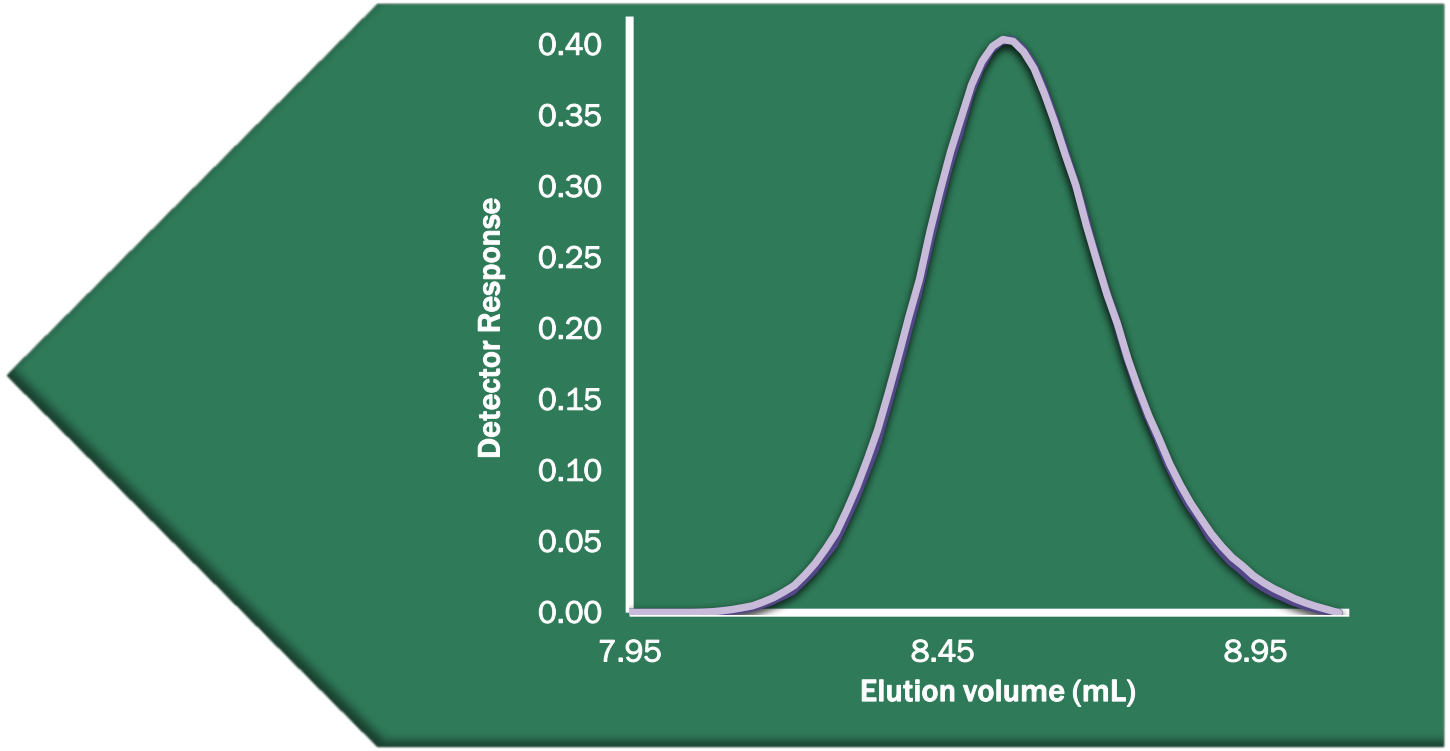
Beyond PEGylated lipids: Non-immunogenic shielding lipids in vaccines

From few grams to kg

R&D batch (100g)
GMP batch (5Kg)





*PSAR has **analogue solubility and macromolecular properties and interactions with water compared to PEG,** with a **wide and versatile C-N terminus group functionality** for **(bio)conjugation** and a proven **GMP manufacturability.***

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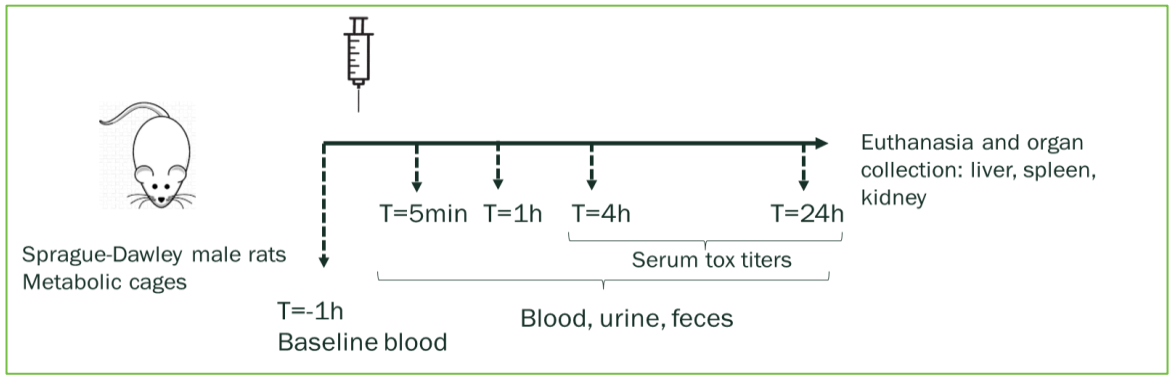
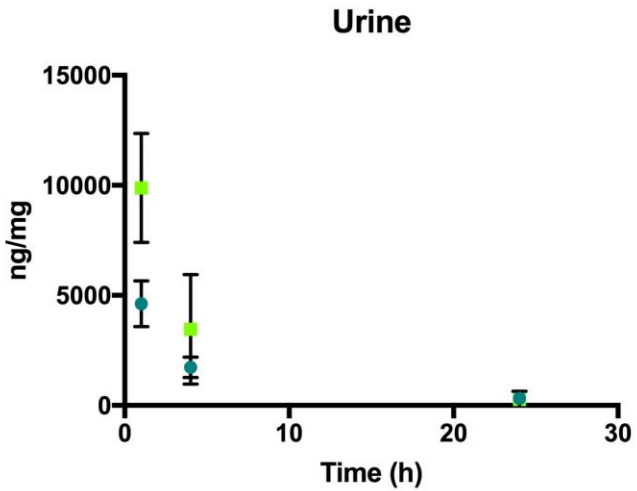
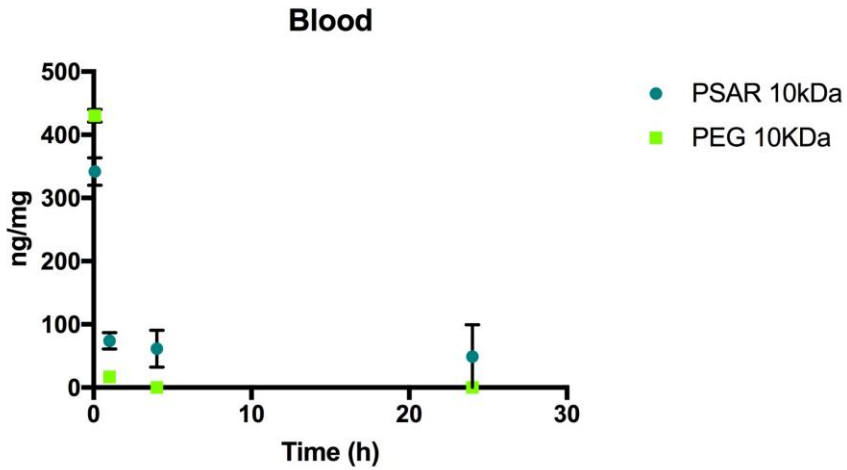
Superior Polysarcosine (Psar) Biological properties

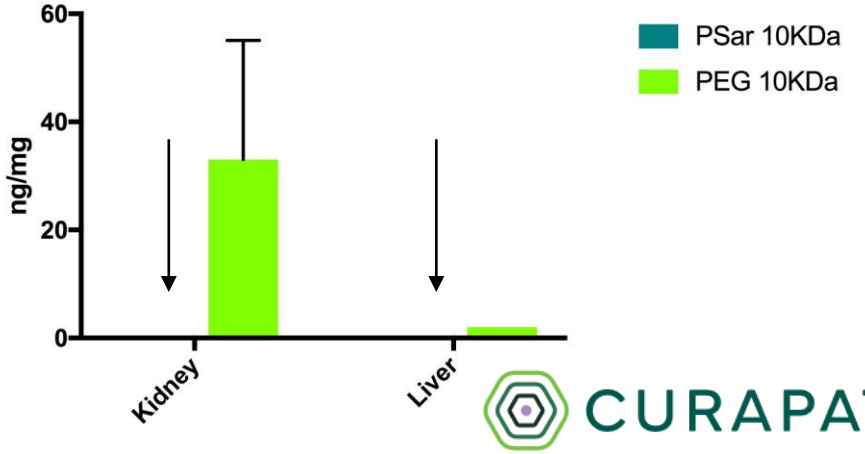
PEG vs PSar Polymer Backbones TOX, Distribution, Elimination Study



- Both PEG and PSar are eliminated through urine.
- Both PEG and PSar Not detected in feces or spleen.
- PEG is detected in Liver and Kidneys
- PSar is not detected in Liver and Kidneys suggesting possible biodegradation







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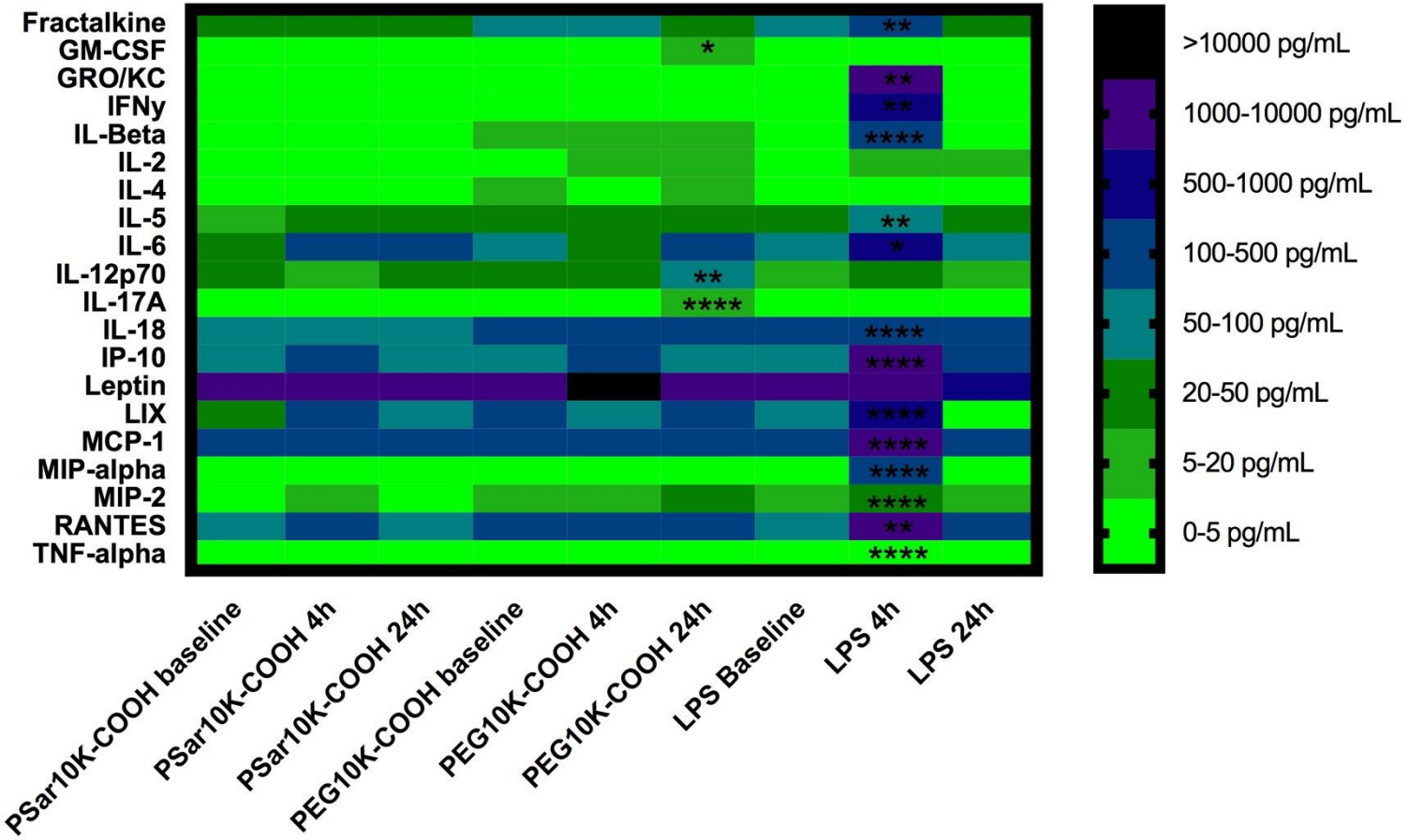
Superior Polysarcosine (Psar) Biological properties

PEG vs PSar Polymer Backbones TOX, Distribution, Elimination Study



- The **MEA-PSAR140-COOH polymer did not induce a systemic immune response since the level of inflammatory mediators stays the same as the baseline**.
- However, there was an increase in few systemic inflammatory mediators 24h **after the administration of MEO-PEG10K-COOH**.
- Remarkably: **IL-17 regulates foreign body response to synthetic materials**

(L. Chung et al. Sci Traslational Med 2020, 12, 539)

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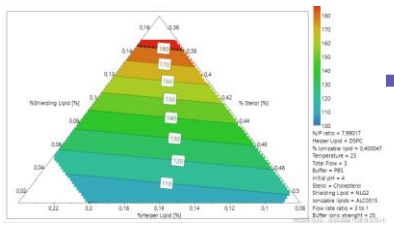
Curapath's shielding lipids in LNP formulations



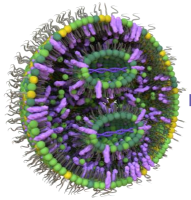
DoE

Automated Pumps

IJM

T-Mixer

Microfluidics



Buffer Exchange
& Concentration

Size and PDI

mRNA Content

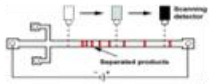
DLS & UV/Vis

Lipid

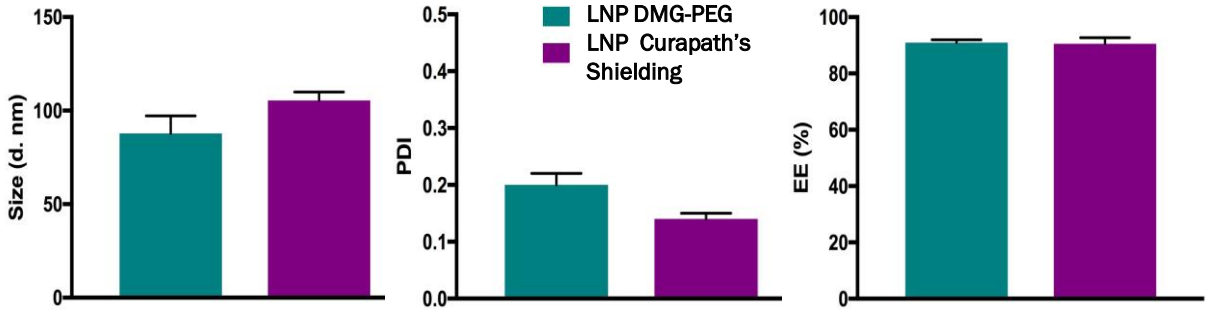
concentration

HPLC CAD

mRNA Integrity



Automated Electrophoresis



*Curapath's shielding lipid yields LNPs with **adequate size in the 100 nm range and low PDIs**, with **>90% nucleic acid** encapsulation efficiency (commercial benchmark, mRNA-Luc)*

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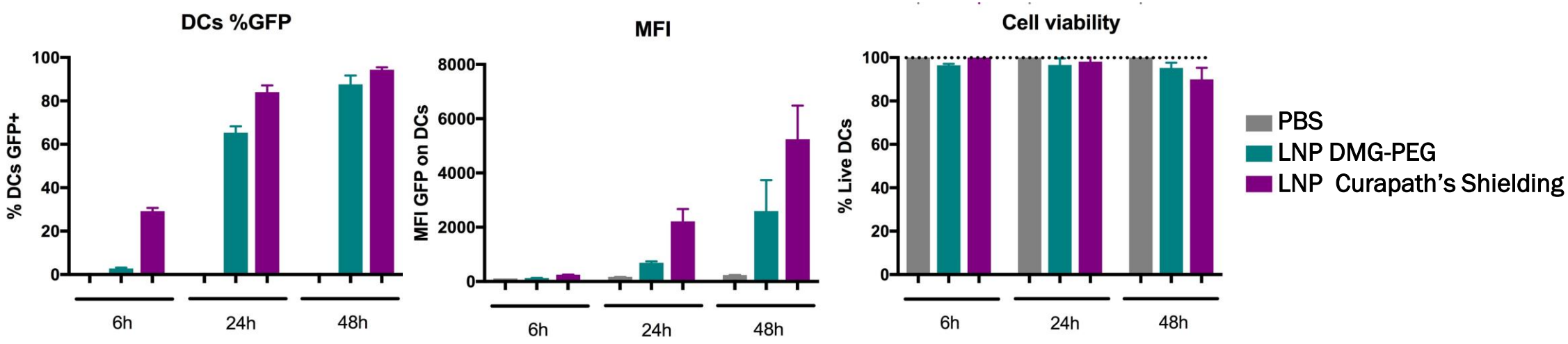
Curapath’s shielding lipids in LNP formulations

In vitro testing



Curapath’s shielding lipid perform equally as Benchmark in terms of transfection efficiency in vitro without affecting cell viability

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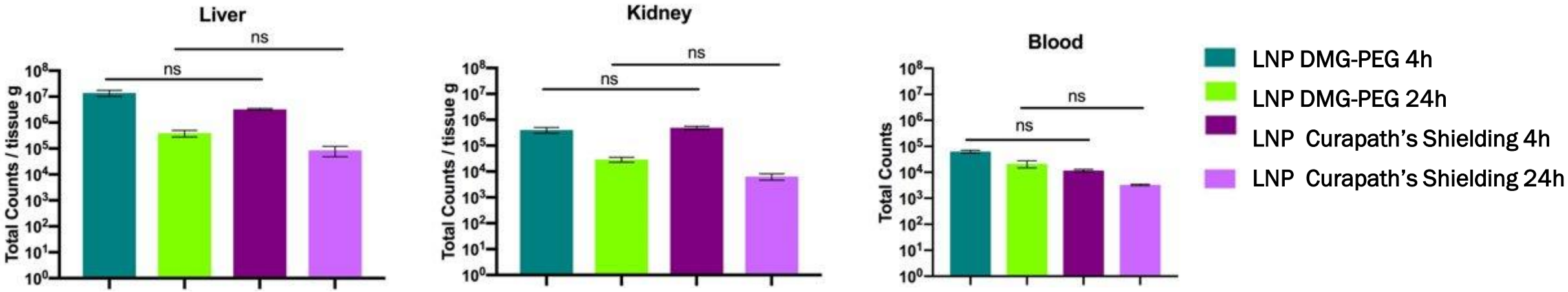
Curapath’s shielding lipids in LNP formulations

In vivo i.v. testing



Curapath’s shielding lipid perform equally as Benchmark in terms of transfection efficiency in vivo without toxic effects

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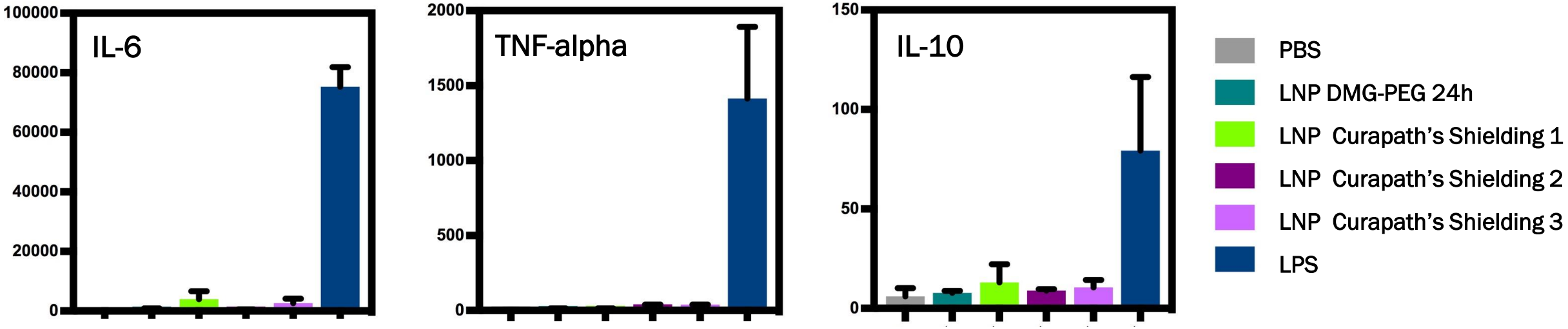
Curapath’s shielding lipids in LNP formulations

In vivo i.v. testing

Pro-inflammatory markers and cytokine levels measured by ELISA in serum samples of Balb/C mice 2h after the i.v. injection



LNPs with proprietary shielding lipid do not induce significantly different release of pro-inflammatory markers and interleukines when compared to Benchmark

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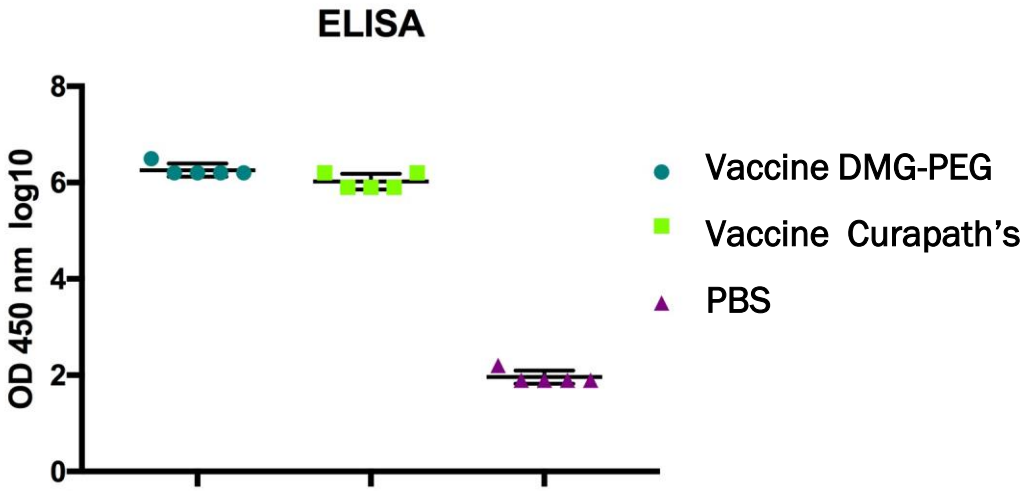
Curapath’s shielding lipids in LNP formulations

In vivo i.m. testing



- *Antibody titers by ELISA against a SARS-CoV-2 relevant protein measured as optical density (OD) at 450 nm at the end point of the experiment, 2 weeks after day 0 and day 14 intramuscular immunizations with Vaccine LNPs with DMG-PEG and Vaccine LNPs with Curapath’s Psar shielding lipid.*

LNPs with proprietary shielding lipid perform equally as Benchmark in terms of immune response in vaccine applications

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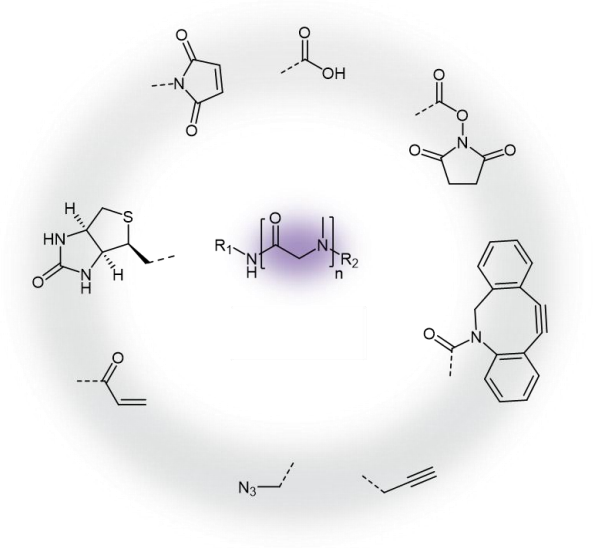
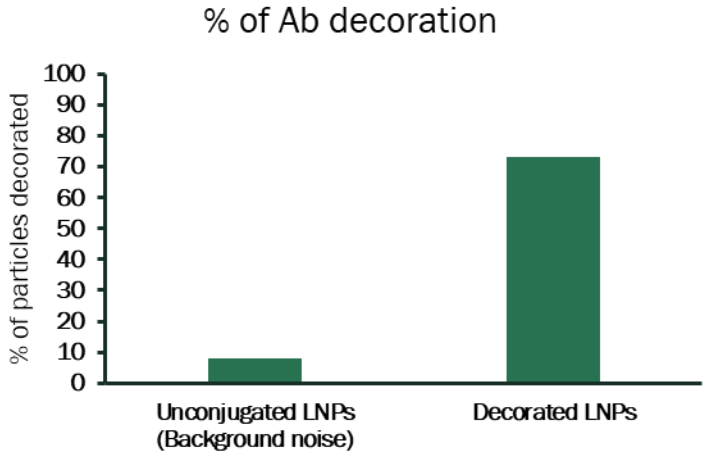
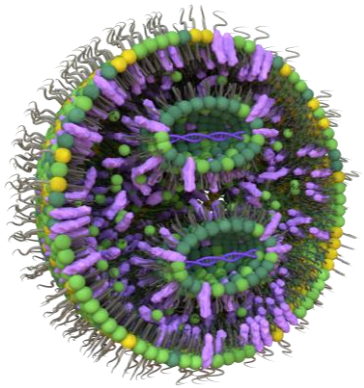
Advancing Precision Delivery: LNP Surface Modification Techniques



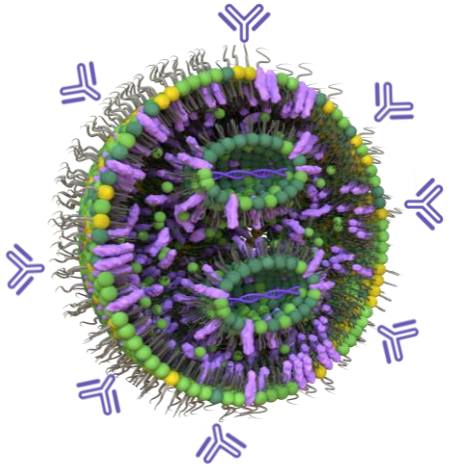
The chemical versatility and robust manufacturing procedure of Psar allows for controlled functionalizations for nanoparticles surface modification → targeting extra-hepatic tissues

Antibody decoration assessment by Cytometry

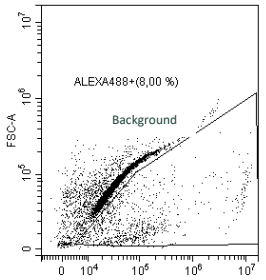




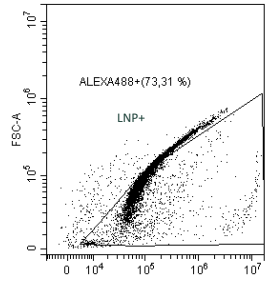
"Efficient – Simple – Clean"
"click-Chemistry"



Undecorated

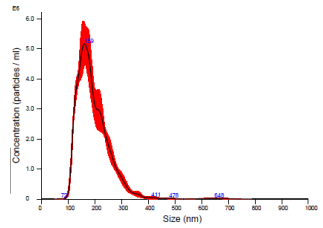


Decorated



Particle concentration

5.89e+008 +/- 1.68e+007 particles/ml



Curapath's shielding lipids opportunities

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Manufacturing feasibility

- Curapath's shielding lipids have been synthesized and scaled-up with batch-to-batch reproducibility.

GMP production

- GMP manufacturing of polymer-lipids in GMP multi-gran scale at Curapath's plant.

Strong patent protection

- Excellent patent position overcoming PEG components.
- Priorities from 2021/2022 & national phases 2024. Expected worldwide protection through 2041/2042.

LNP formulations

- Curapath's shielding lipid yield NPs with adequate size in the 100 nm range and low PDIs, with >90% nucleic acid encapsulation efficiency.

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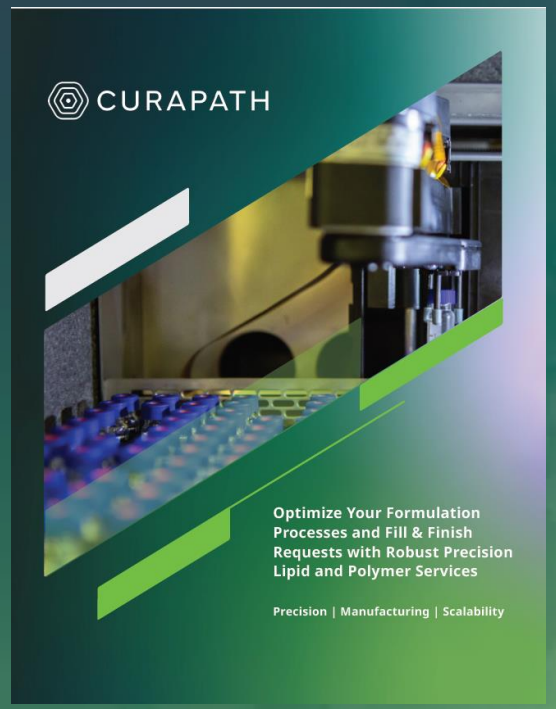
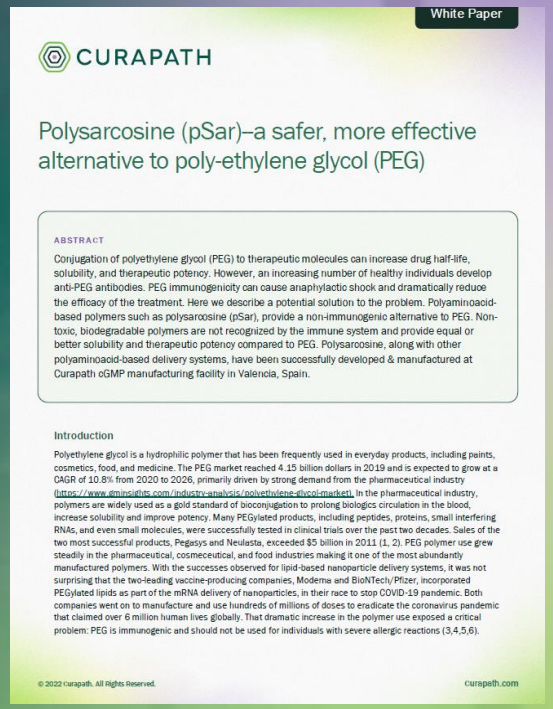
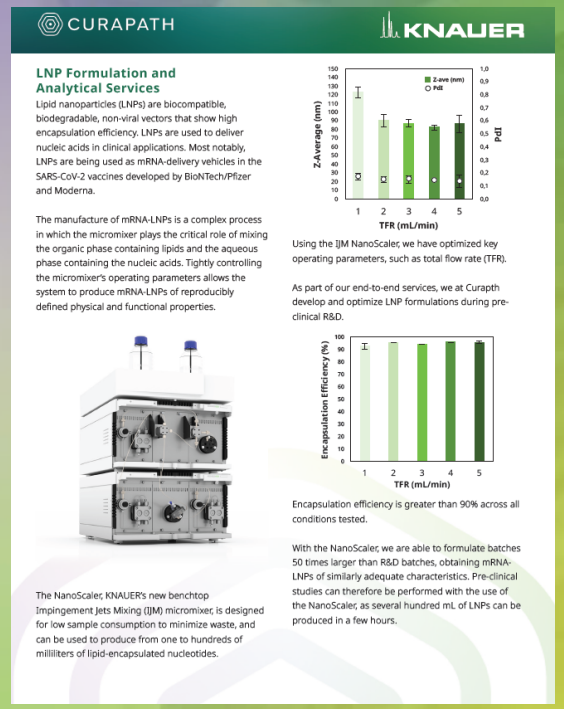
Brochures, Product Catalogue, Case Studies, Product Highlights, App Notes, Posters…..







… and more to come

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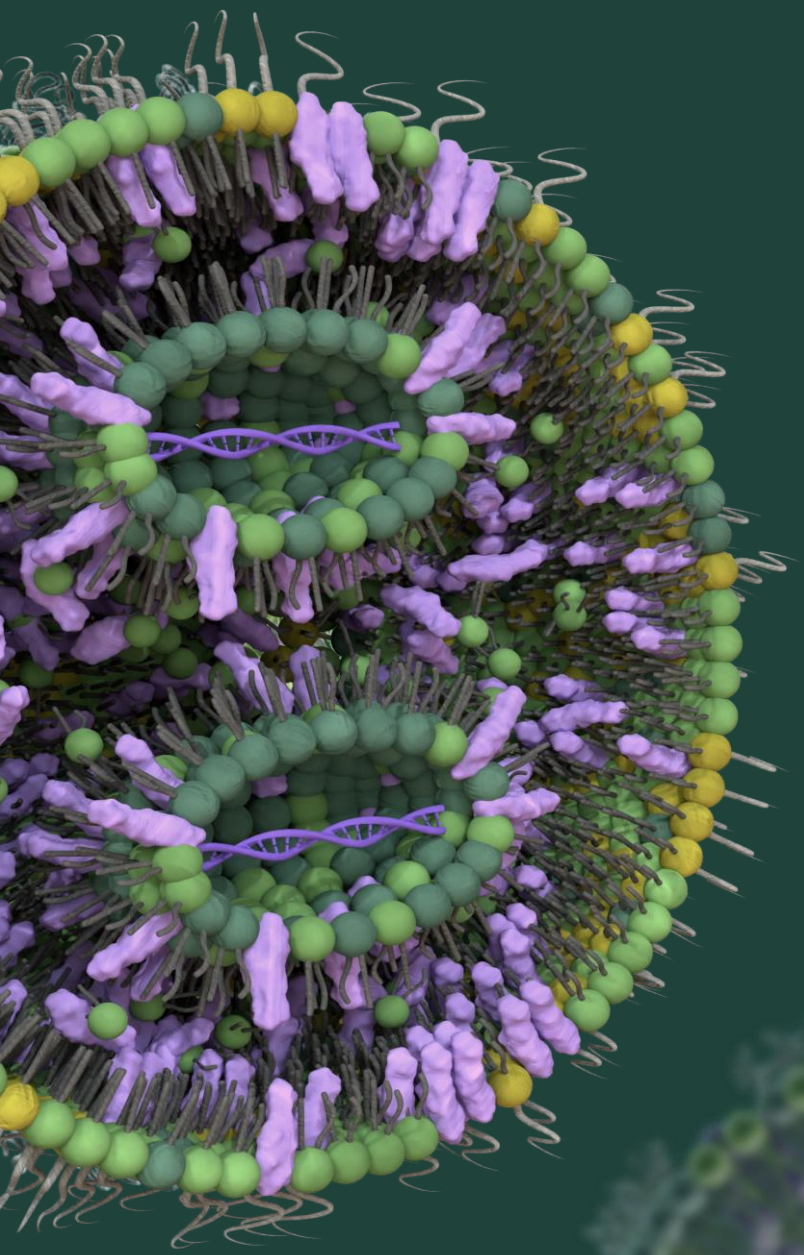
Booth #61

www.curapath.com



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GMP production site

Av. Benjamín Franklin, 19
46980 Paterna, Valencia

Innovation & Development site

Carrer d'Alexander Graham Bell, 6
46980 Paterna, Valencia

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