university of groningen / faculty of science and engineering

Understanding how the physico-chemical properties of lipid-based drug carriers shape their interactions in complex biological environments

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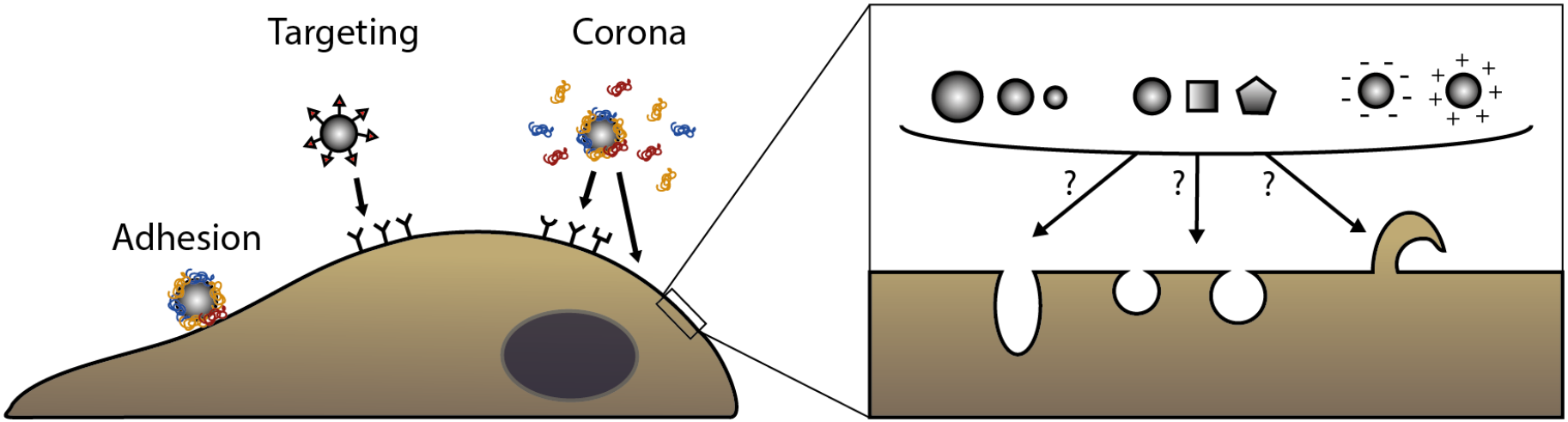
Nanomedicine for drug delivery

Trying to understand how nanomedicines interact with cells and how to control it

→ "the cell biology of nanomedicine"

1) Interactions at the cell membrane (receptors and targeting)

2) Uptake and intracellular processing



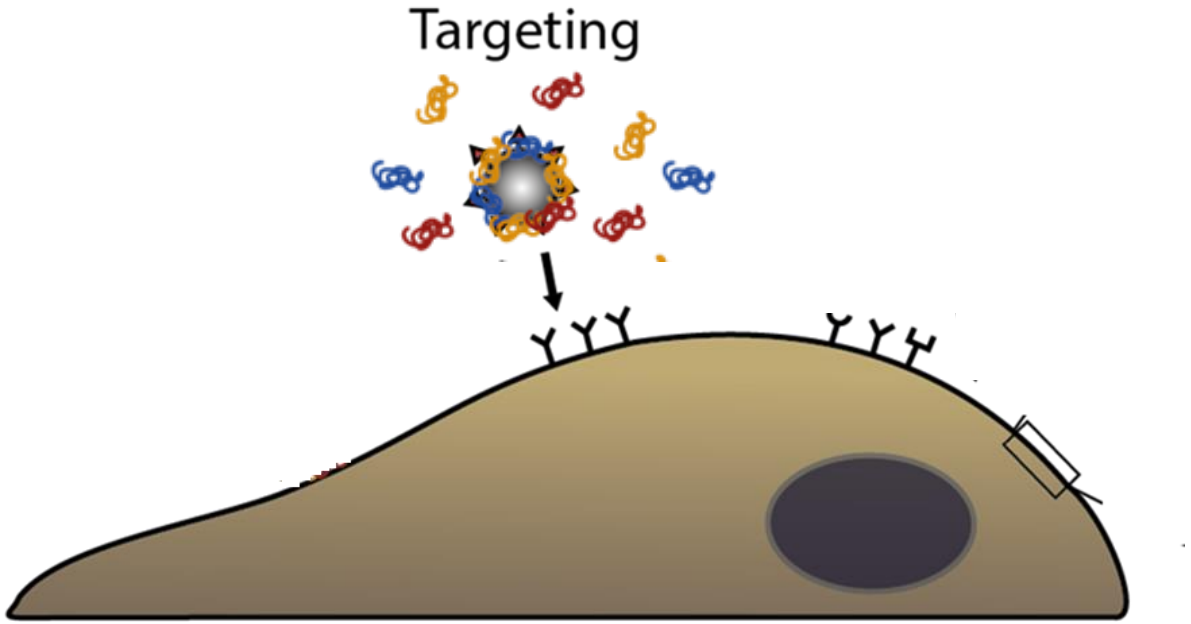
We need to learn the language to make it better!

How do nanomedicine physico-chemical properties shape their biological behavior?

Francia et al, ***Beilstein J Nanotech****, 2020, 11 (1), 338-353*
Interactions at the cell membrane and pathways of internalization of nano-sized materials for nanomedicine

Can we control the interaction with cell receptors?

- Changing nanomedicine physico-chemical properties
- Active targeting ligands on nanomedicines to interact with specific cells



but upon administration: interactions with biological fluids and corona formation
→ *BEFORE* interactions with cells!

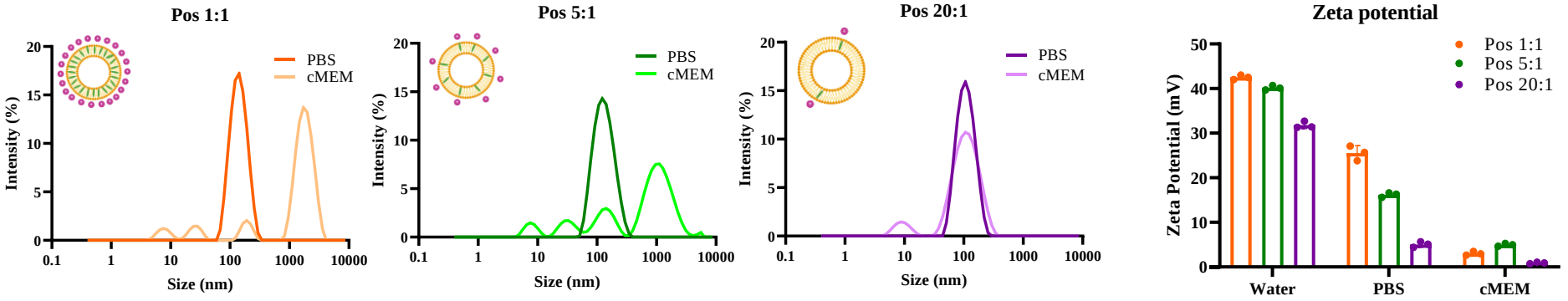
What are the implications for the interactions with cells?
…some examples

Why do positive nanoparticles usually have higher uptake than negative ones?

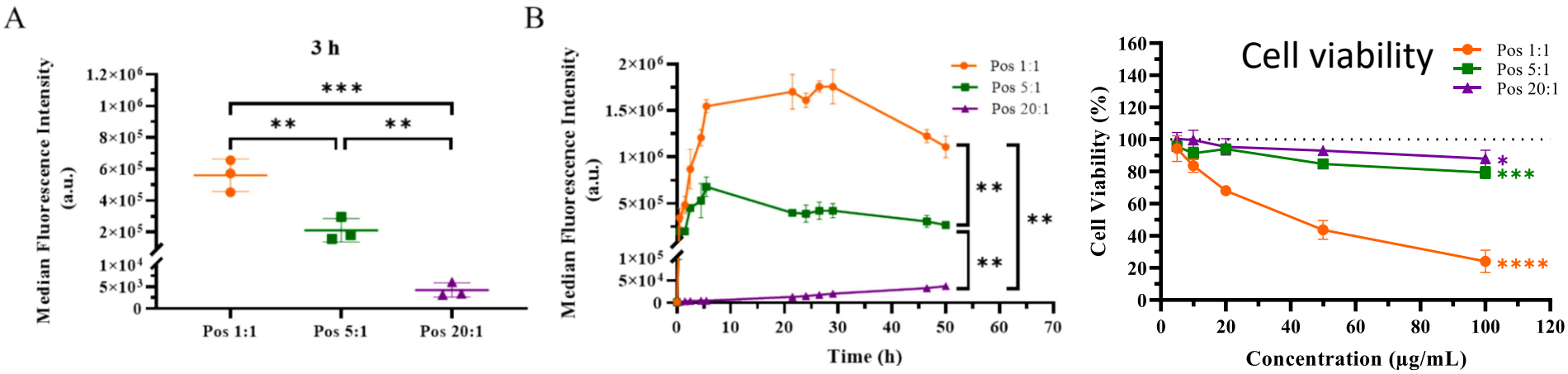
Feng Zhao

Optimization of DOPC-Dc-Chol liposome formulation

→ ***to find a non-toxic and stable positive liposome***



Pos 1:1 and 5:1 aggregate in the medium with 10% FBS, 20:1 is stable

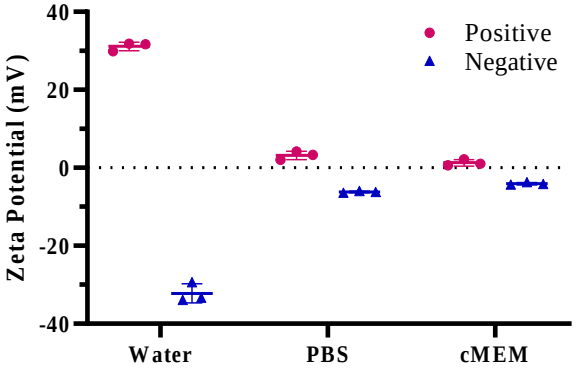


Feng Zhao, Anna Salvati, et al. **INT J PHARMACEUT**, 681, 125881, 2025.
Effect of positive charges on liposome stability, uptake efficacy and impact on cells

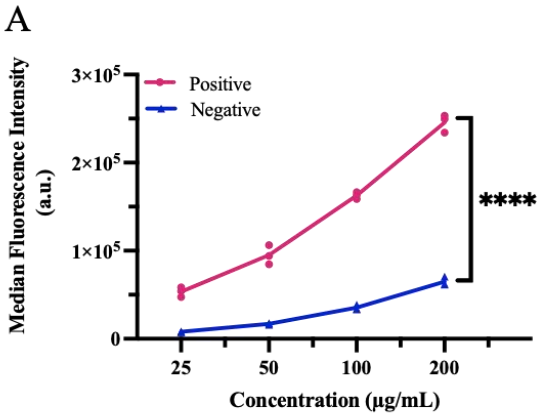
Why do positive nanoparticles usually have higher uptake than negative ones?

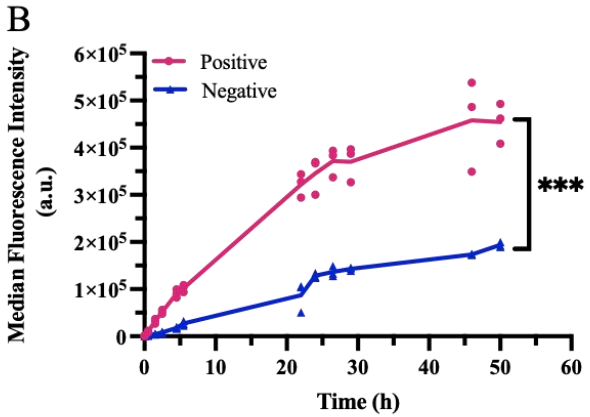
Feng Zhao

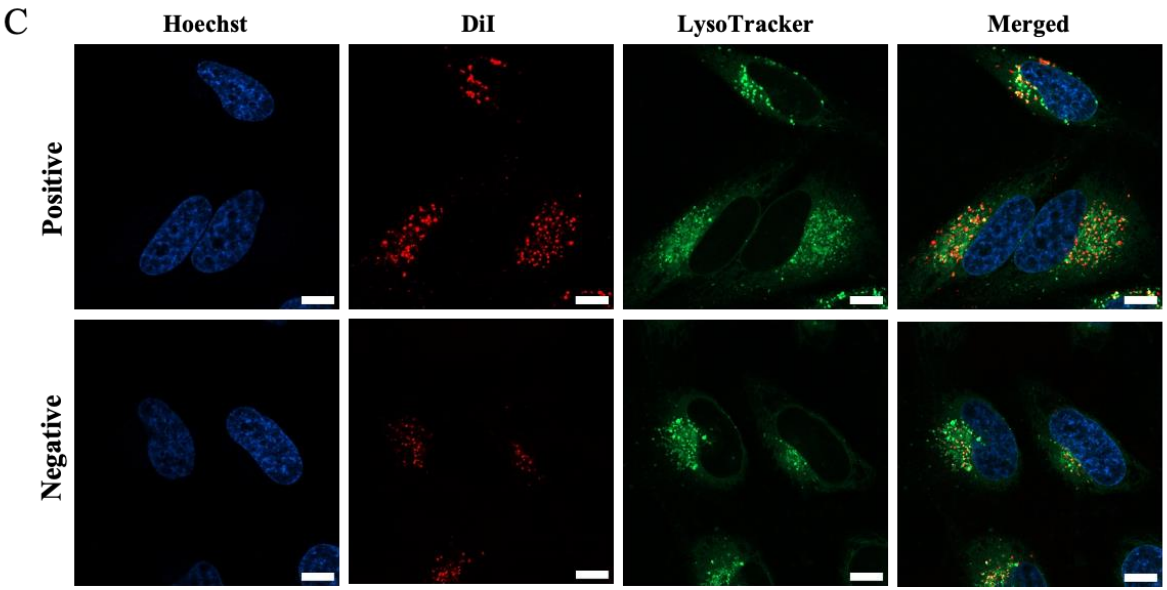
Preparing a corresponding liposome with the same amount of charges but negative



Both are near neutral in serum





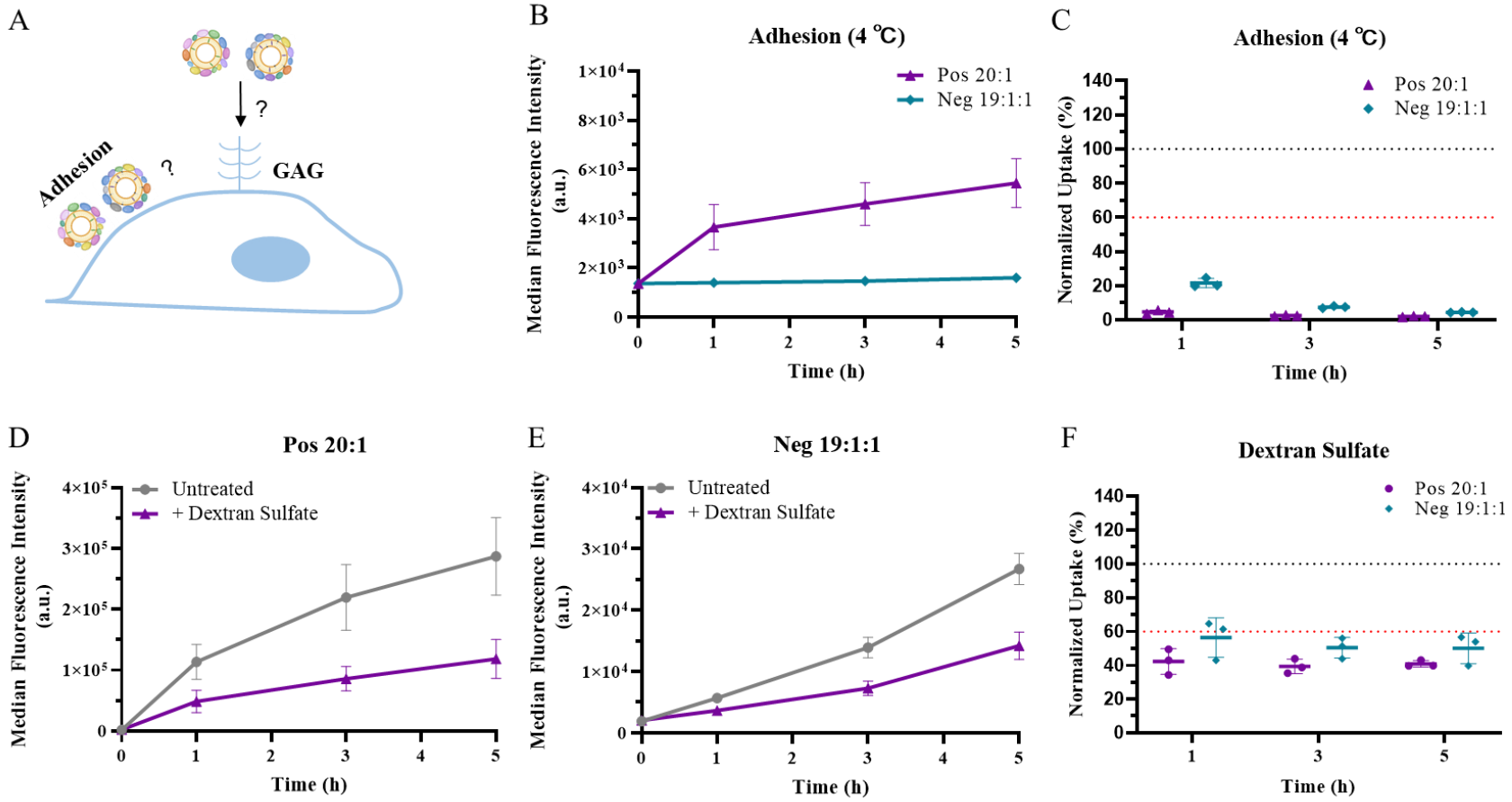


Feng Zhao, Anna Salvati, et al. **INT J PHARMACEUT**, 681, 125881, 2025.
Effect of positive charges on liposome stability, uptake efficacy and impact on cells

Why do positive nanoparticles usually have higher uptake than negative ones?

Feng Zhao

Common explanation: interaction with negatively charged proteoglycans on the cell membrane



→ *Instead they both interact with heparan sulphate for uptake!*

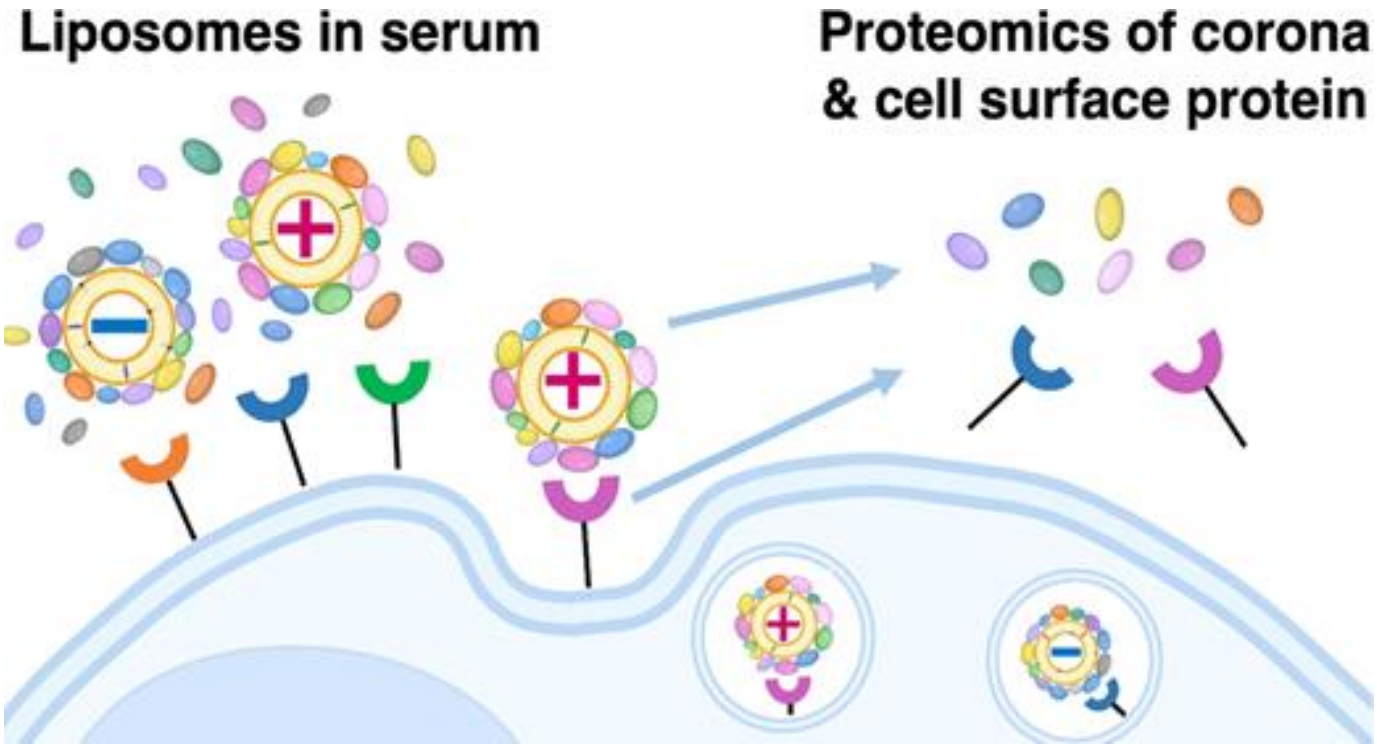
→ *Slightly higher adhesion to cell membrane for positive liposomes*

Feng Zhao, Anna Salvati, et al. **INT J PHARMACEUT**, 681, 125881, 2025.
Effect of positive charges on liposome stability, uptake efficacy and impact on cells

Comparison of Protein Coronas and Internalized Cell Surface Proteins of Positive and Negative Liposomes

Feng Zhao

1. Protein corona adsorbed on the liposomes
2. Cell surface proteins involved in their internalization.



Feng Zhao, Anna Salvati, et al. **ACS Nano,** 2026, **20 (27): 19225–19240**
Comparison of Protein Coronas and Internalized Cell Surface Proteins of Positive and Negative Liposomes

Comparison of Protein Coronas and Internalized Cell Surface Proteins of Positive and Negative Liposomes

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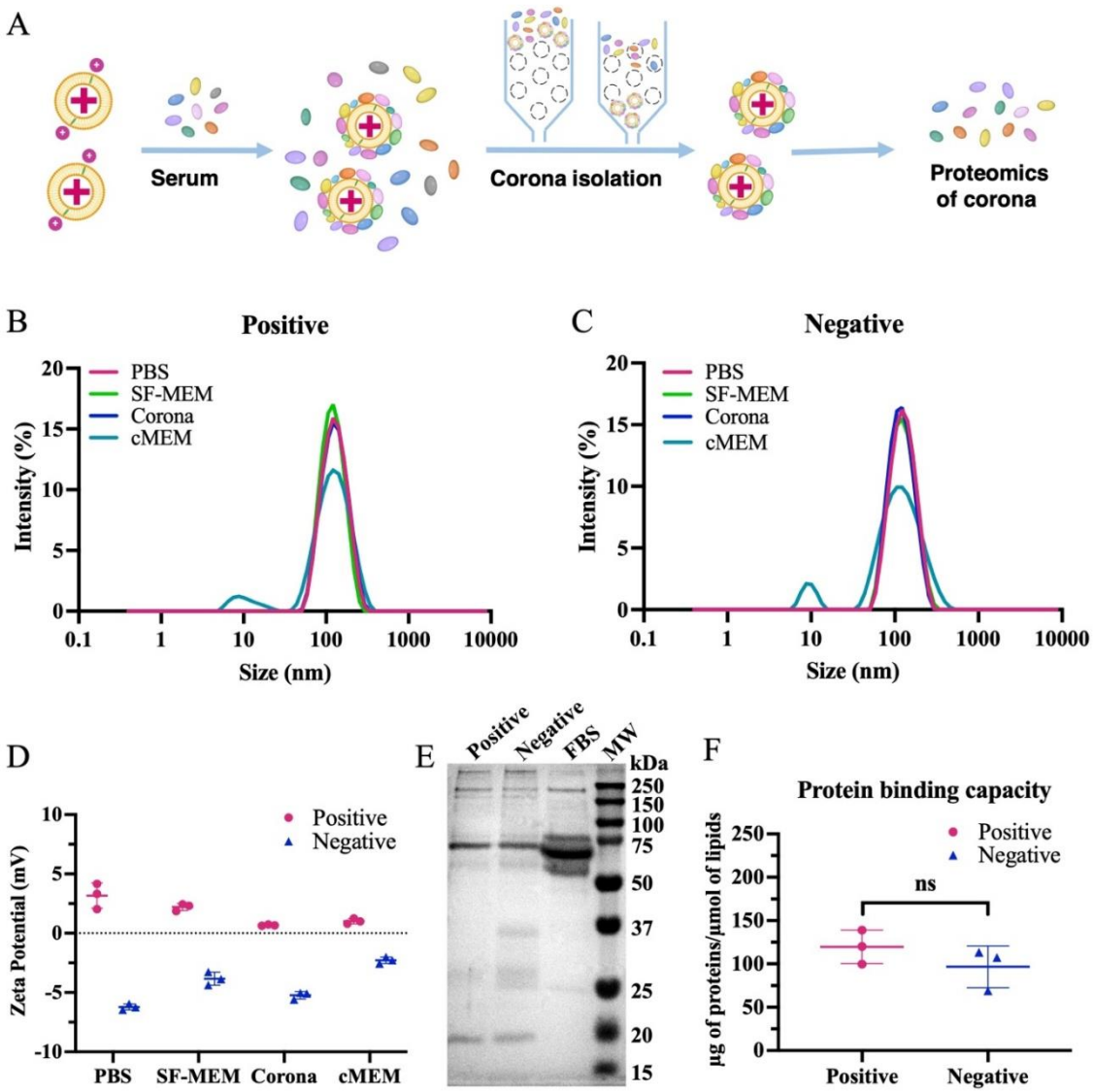

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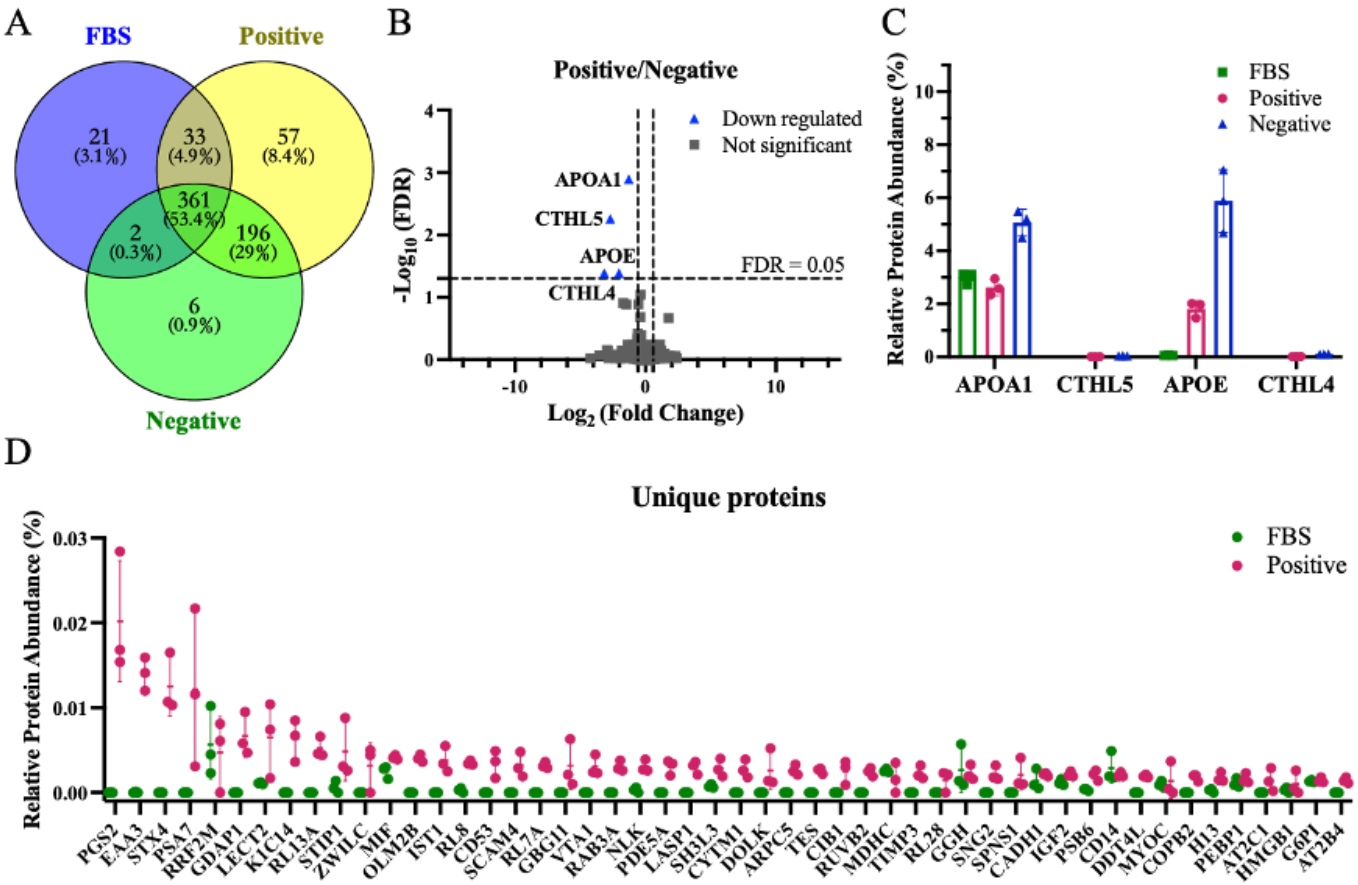
Comparison of Protein Coronas and Internalized Cell Surface Proteins of Positive and Negative Liposomes

Feng Zhao

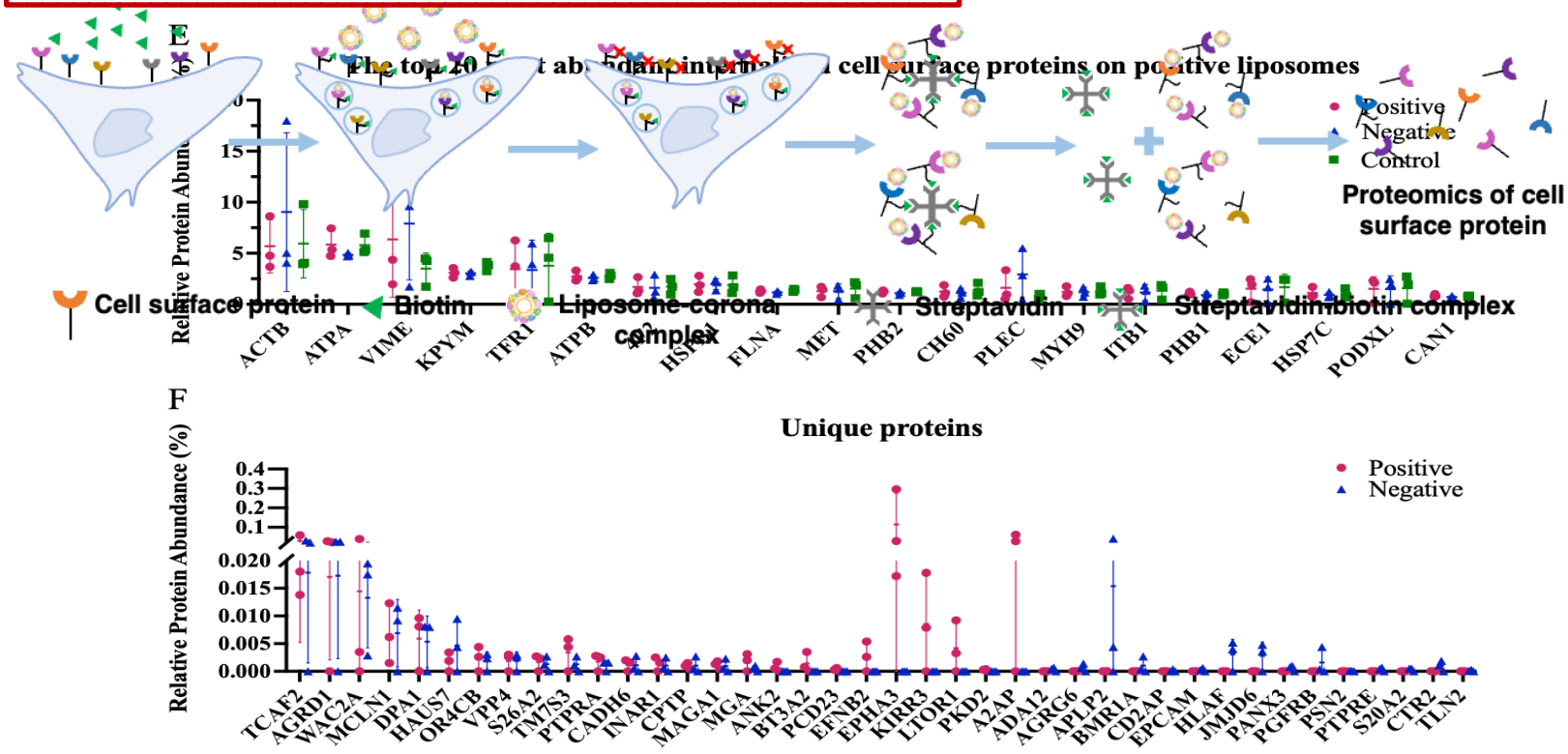
- Major differentially expressed proteins have low abundance
- Many of the most abundant proteins are common but differed in abundance
- ***NB: not always few dominating corona proteins!***



Feng Zhao, Anna Salvati, et al. **ACS Nano,** 2026, **20 (27): 19225–19240**
Comparison of Protein Coronas and Internalized Cell Surface Proteins of Positive and Negative Liposomes

Isolation of cell receptors using reversible cell surface biotinylation

Aldy Aliyandi, Anna Salvati, et al **Acta Biomaterialia**, 155, 507-520, 2022



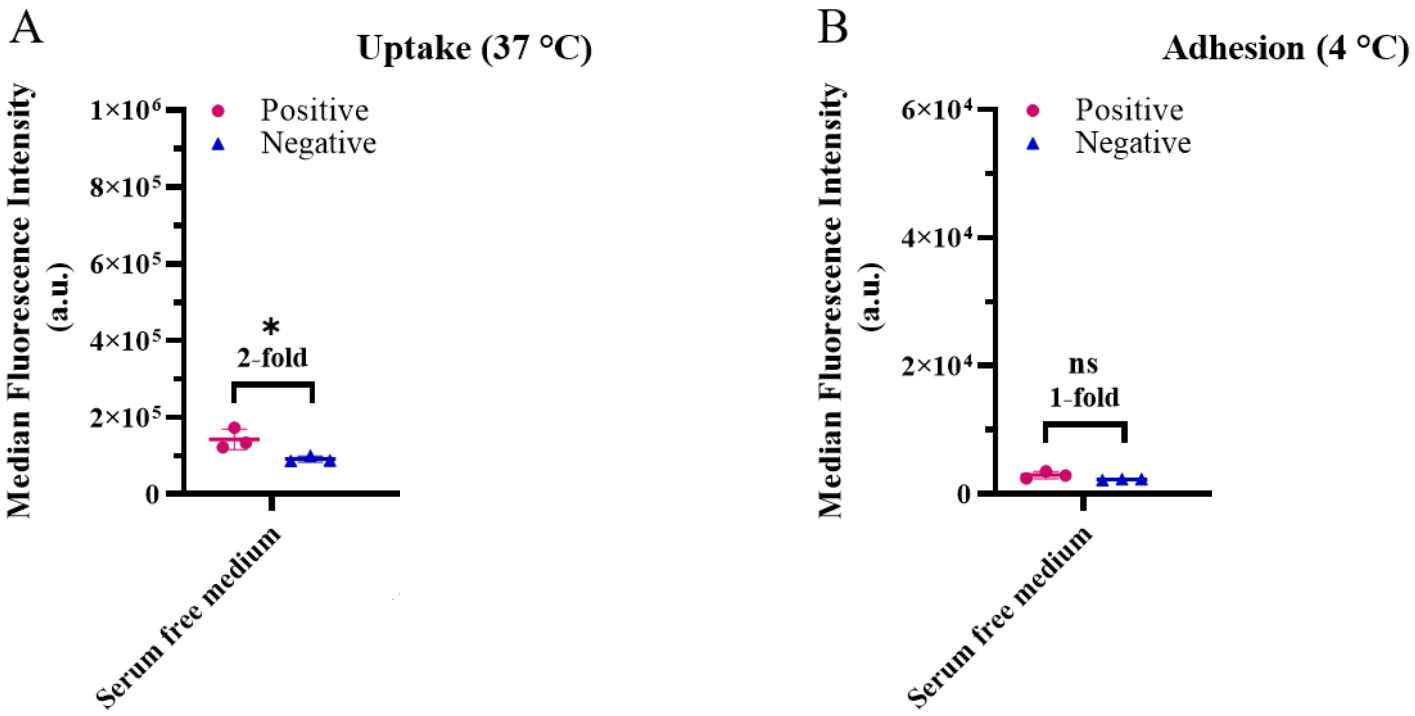
- *Major differentially expressed proteins but low abundant.*
- *Most abundant proteins common in both systems with small differences*
- ***Not few dominating receptors! Much more complex pattern of subtle differences!***

Feng Zhao, Anna Salvati, et al. **ACS Nano,** 2026, **20 (27): 19225–19240**
Comparison of Protein Coronas and Internalized Cell Surface Proteins of Positive and Negative Liposomes

Comparison of Protein Coronas and Internalized Cell Surface Proteins of Positive and Negative Liposomes

Feng Zhao

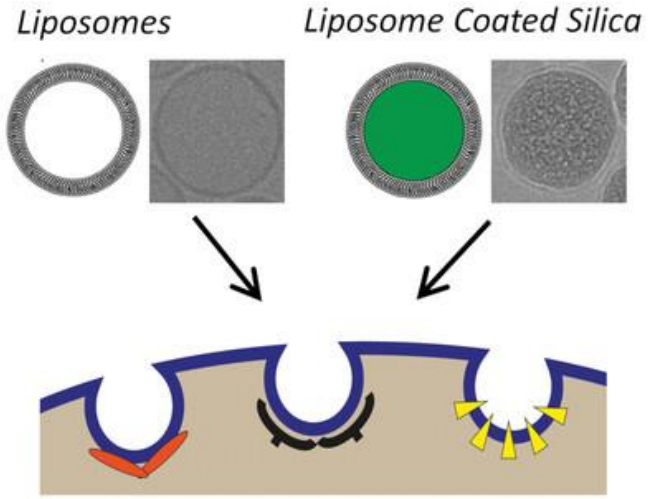
….does it matter?



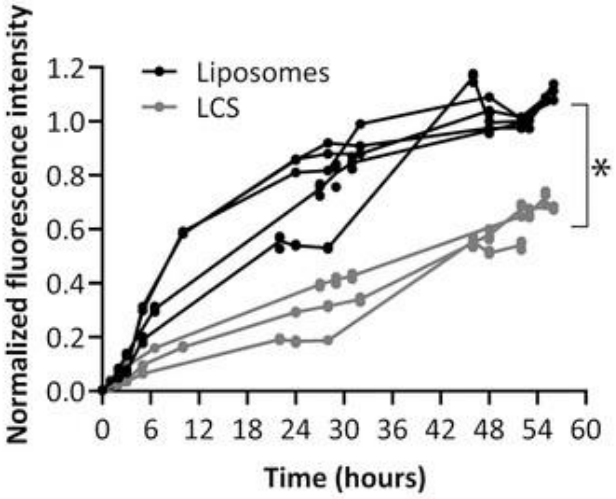
Electrostatic interactions alone cannot explain liposome uptake differences.
The uptake (and adhesion) of two liposomes is different only when proteins are present.

Feng Zhao, Anna Salvati, et al. **ACS Nano,** 2026, **20 (27): 19225–19240**
Comparison of Protein Coronas and Internalized Cell Surface Proteins of Positive and Negative Liposomes

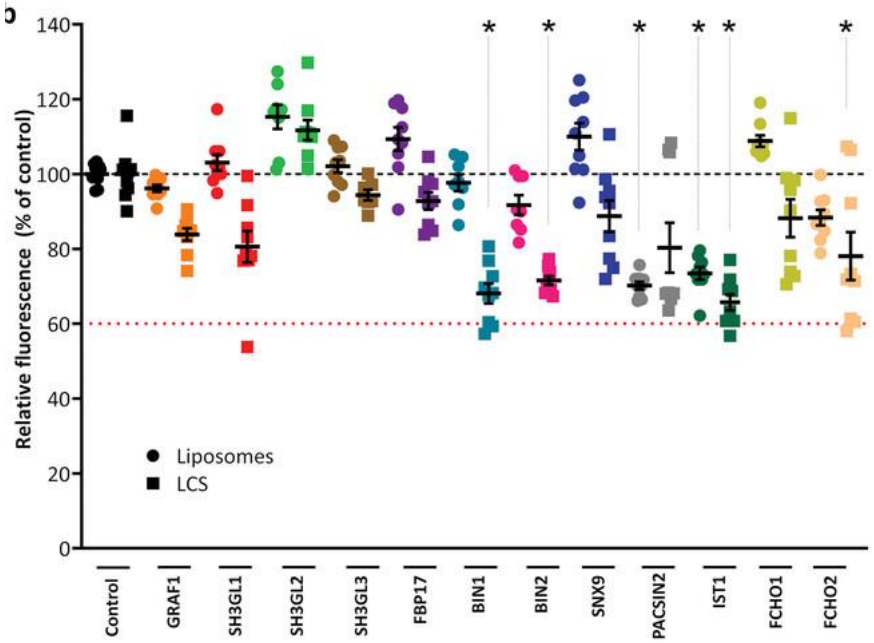
….another example: nanoparticle rigidity



Cell uptake & curvature-sensing proteins?



- Liposomes and liposome coated silica as a model
- Effect on uptake
- Involvement of specialized curvature sensing proteins (BAR domain proteins) in the internalization



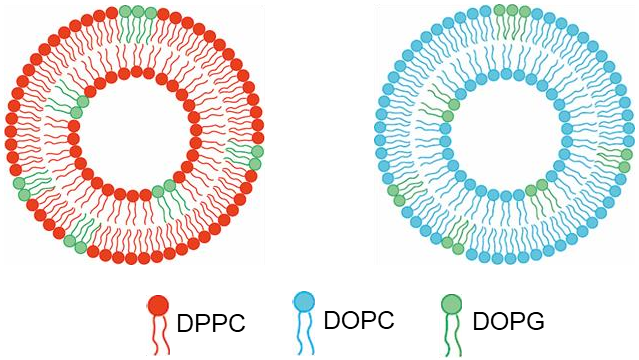
… but does rigidity affect corona composition as well?

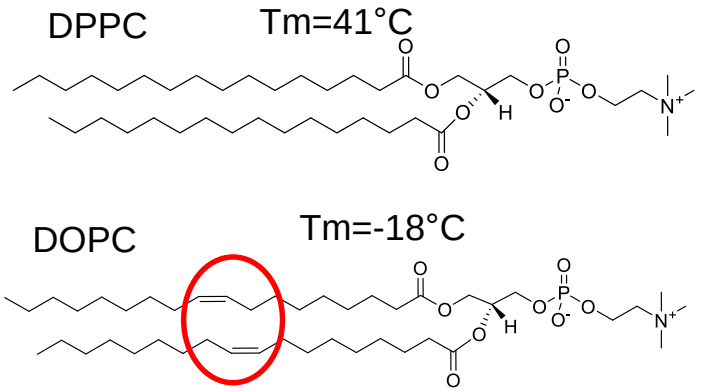
D. Montizaan et al. ***Small***, 2023, 19(39), 2303267
Role of Curvature-Sensing Proteins in the Uptake of Nanoparticles with Different Mechanical Properties.

Changing lipids to tune liposome rigidity

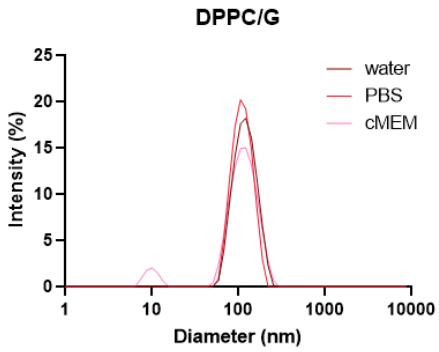
Xinyu Ma

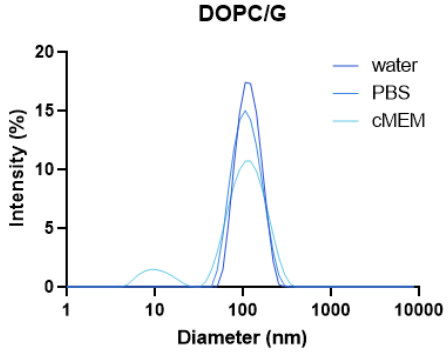
DPPC/G DOPC/G



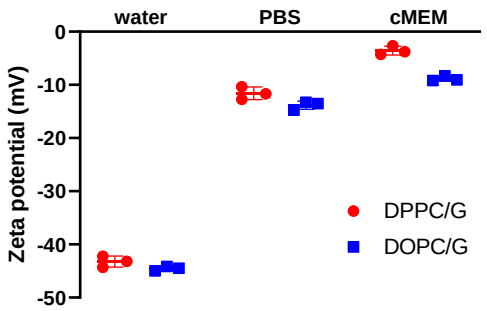


Adding as little as possible DOPG in both formulations to achieve stability





(cMEM=cell medium with 10% FBS)

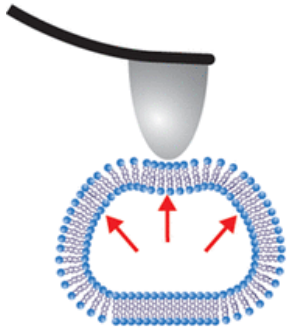


- Extensive optimization needed to obtain non-pegylated liposomes with solid bilayer that were stable in serum!
- Same size, same zeta potential, same surface composition, only acyl chain different!

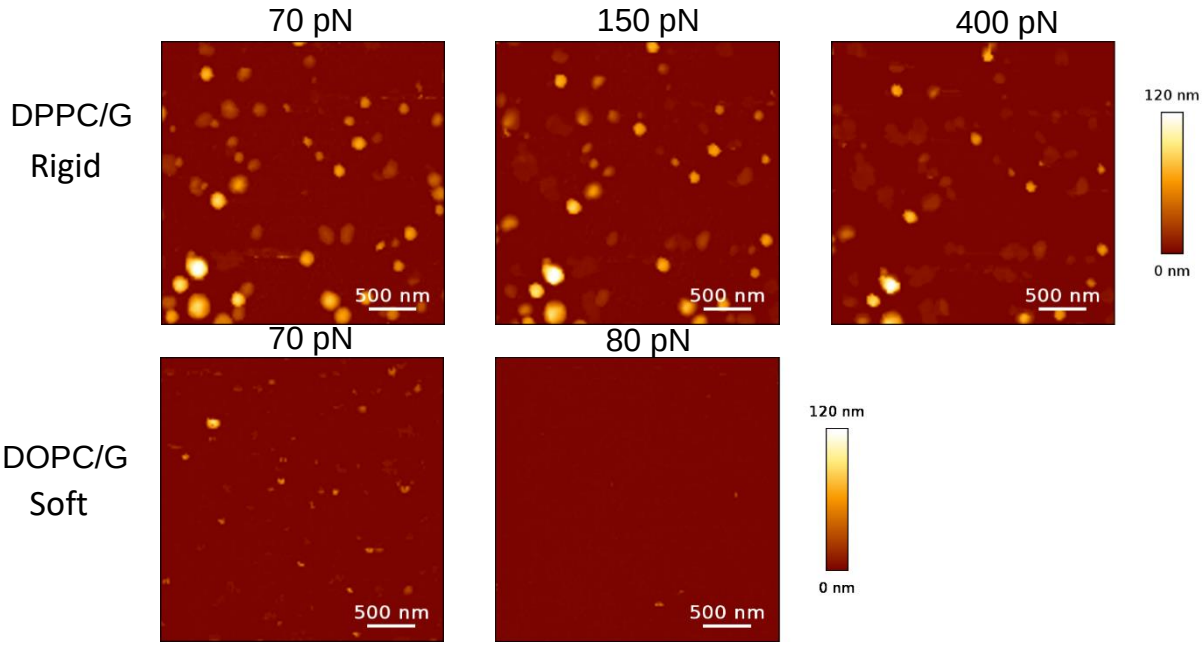
X. Ma et al *Effect of liposome rigidity on corona formation and its implications for uptake by cells.* ***In press 2026***

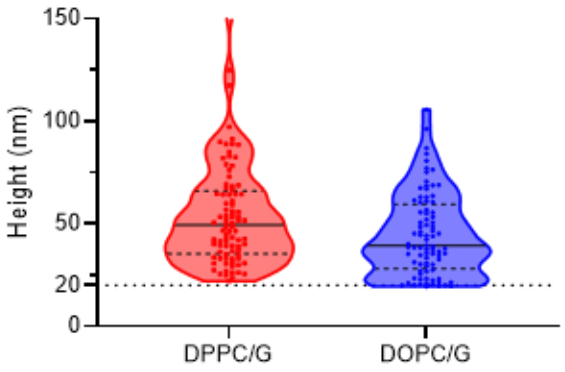
AFM imaging at increasing imaging force

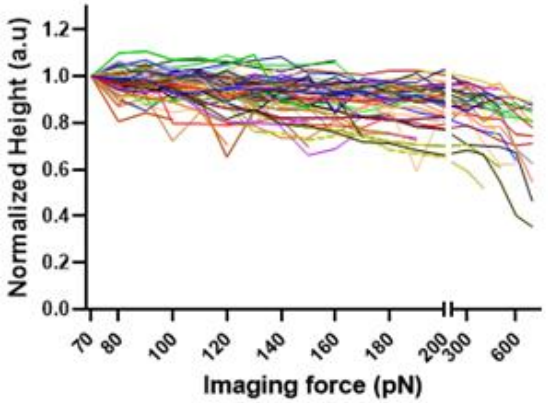
Xinyu Ma

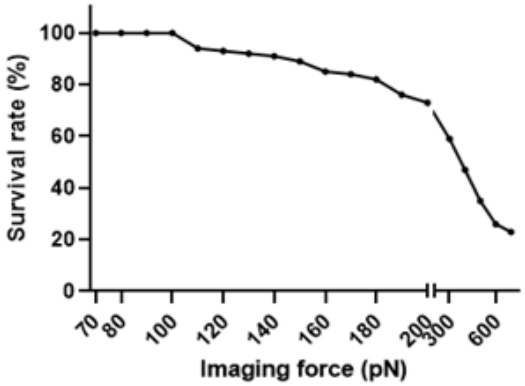


With Wouter H. Roos, Zernike Institute for Advanced Materials (ZIAM)





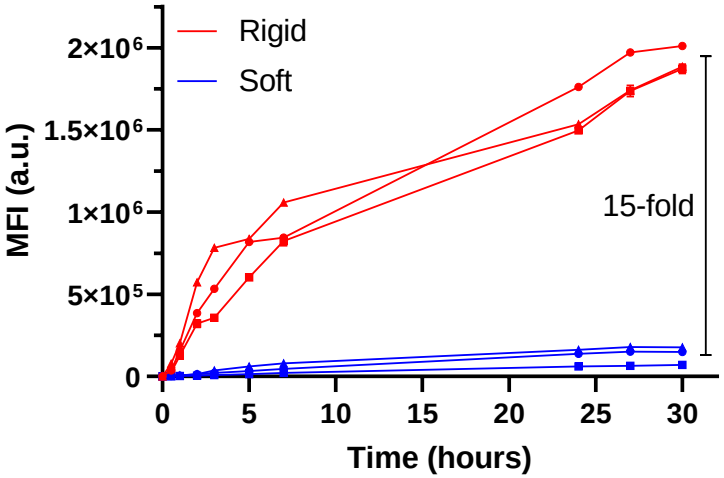


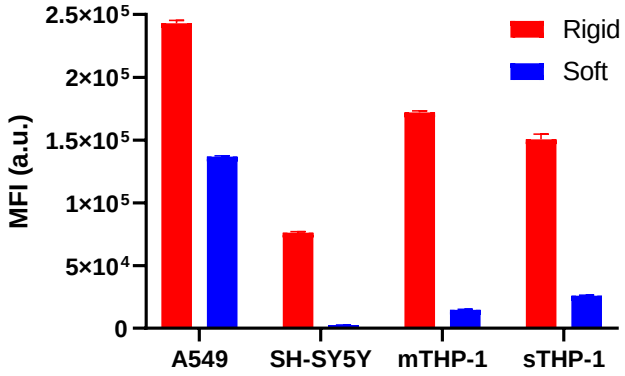


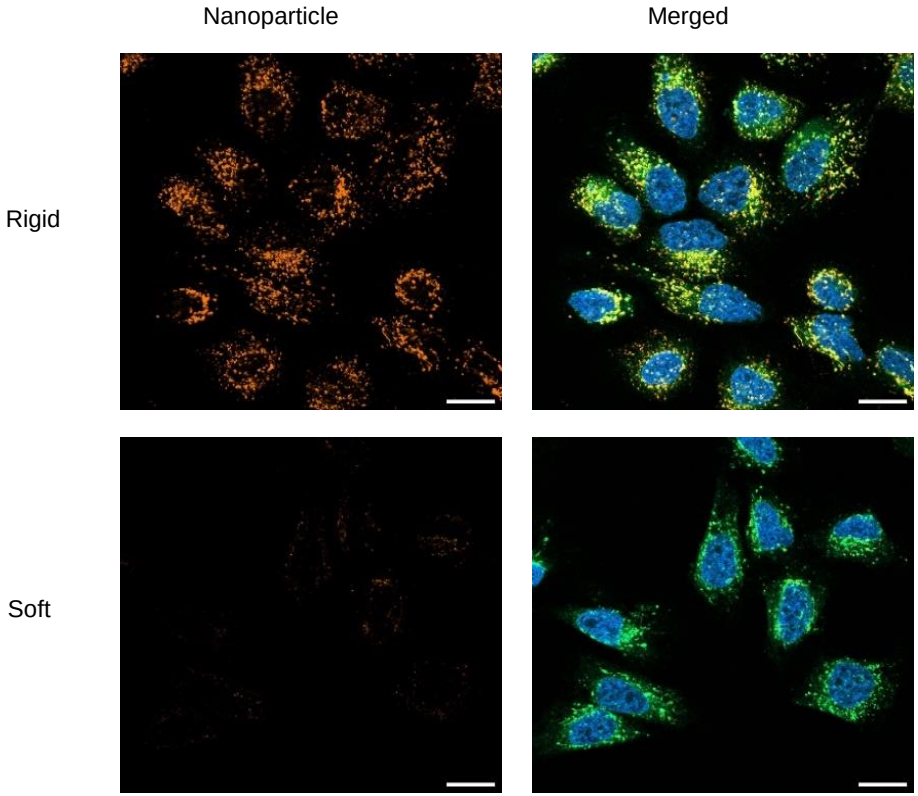
X. Ma et al *Effect of liposome rigidity on corona formation and its implications for uptake by cells.* ***In press 2026***

Rigid liposomes show higher uptake than soft liposomes

Xinyu Ma



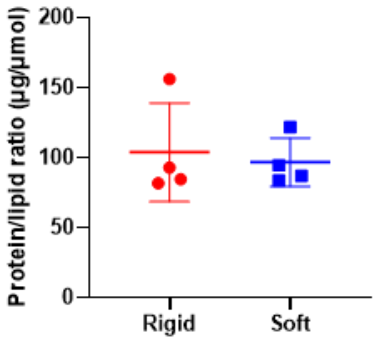


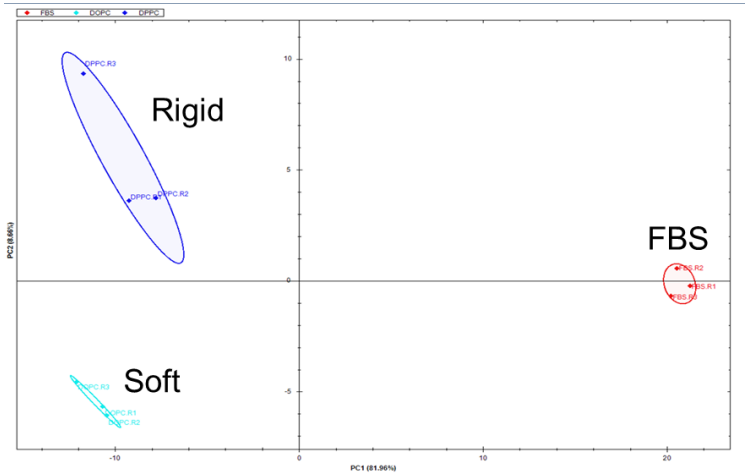


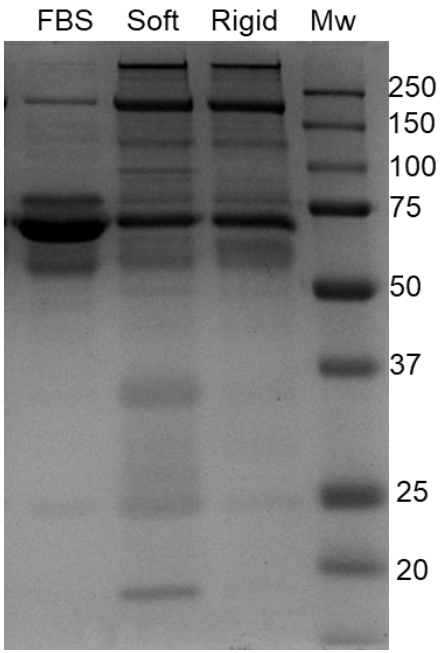
X. Ma et al *Effect of liposome rigidity on corona formation and its implications for uptake by cells.* ***In press 2026***

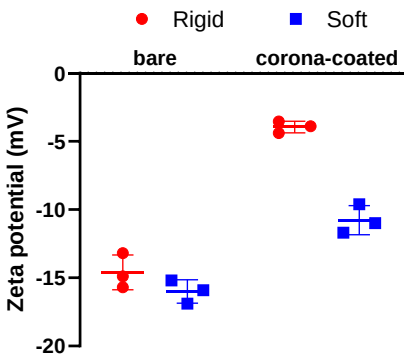
Does NP rigidity affect corona formation as well?

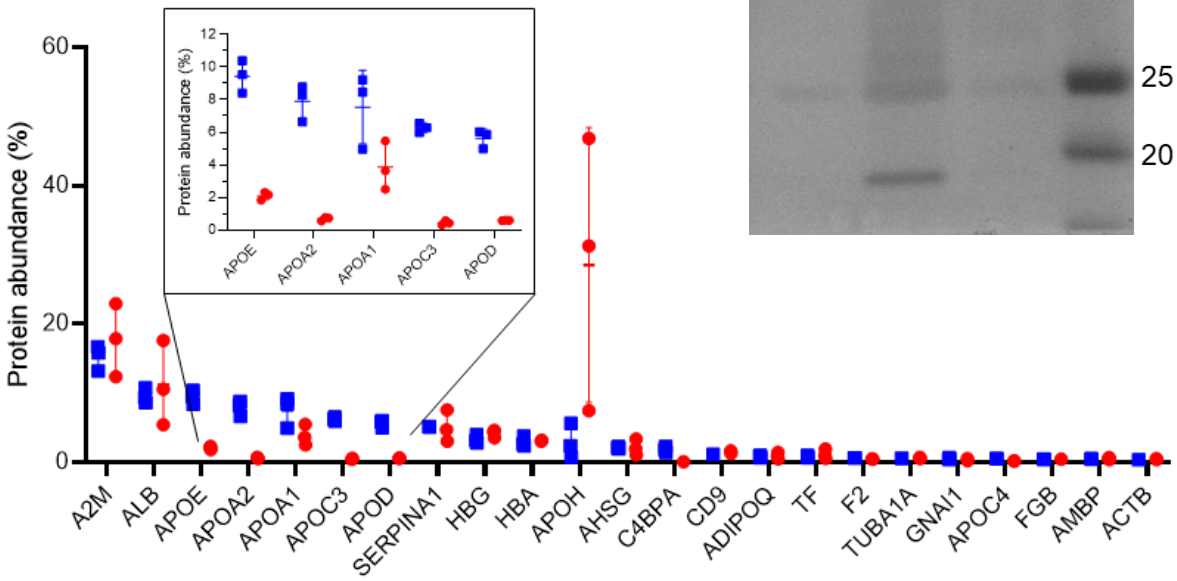
Xinyu Ma











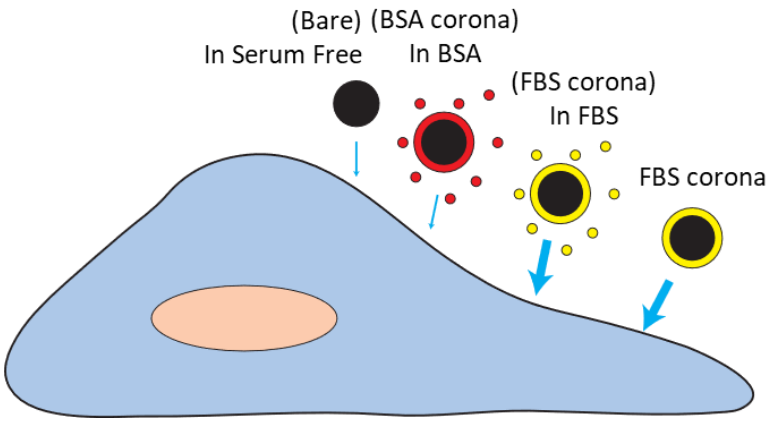
- ***Very different corona!***
- Many common proteins shared
- Many apolipoproteins enriched but different abundance

X. Ma et al *Effect of liposome rigidity on corona formation and its implications for uptake by cells.* ***In press 2026***

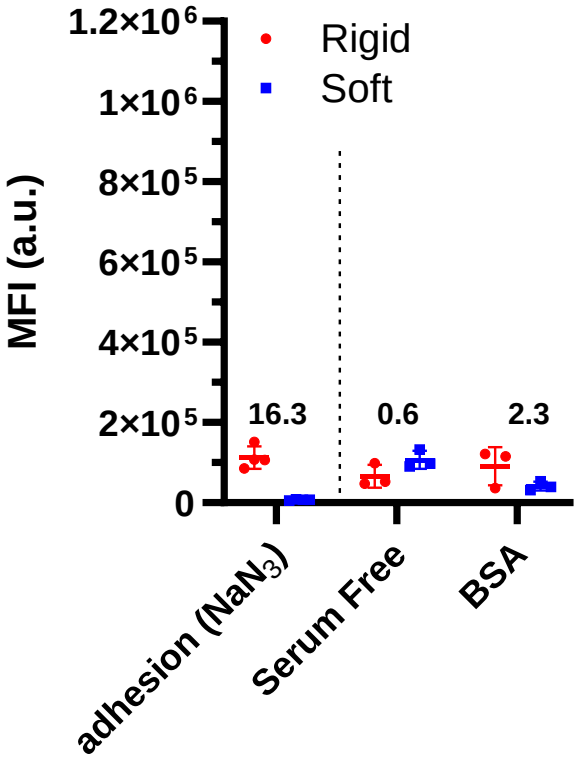
How much does the corona layer matter?

Xinyu Ma

Adding liposomes to cells in medium with different proteins



in different media



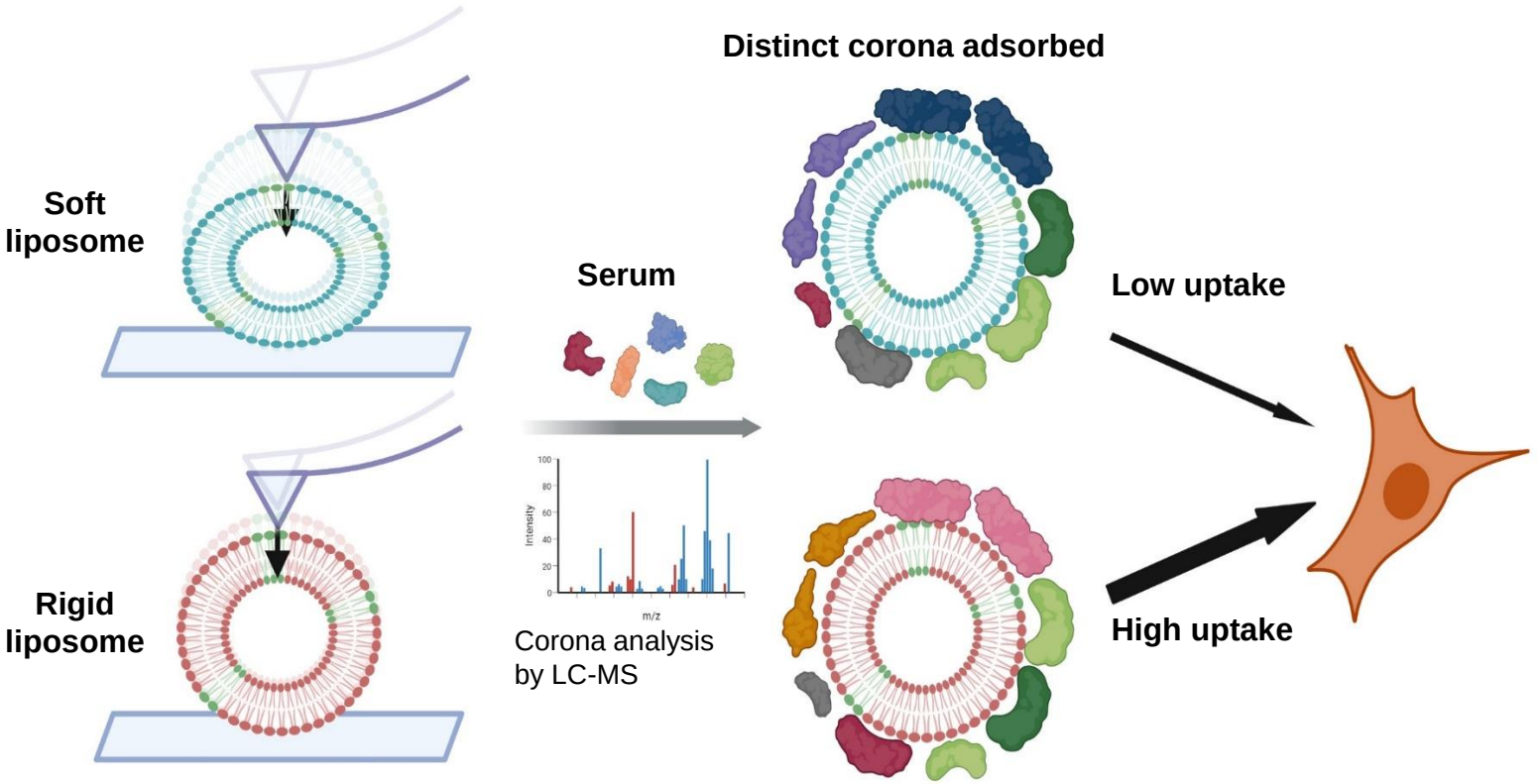
Only when adding liposomes to cells with serum, uptake is different!

X. Ma et al *Effect of liposome rigidity on corona formation and its implications for uptake by cells.* ***In press 2026***

Effect of liposome rigidity on corona formation and its implications for uptake by cells

Xinyu Ma

Xinyu Ma, see poster #25

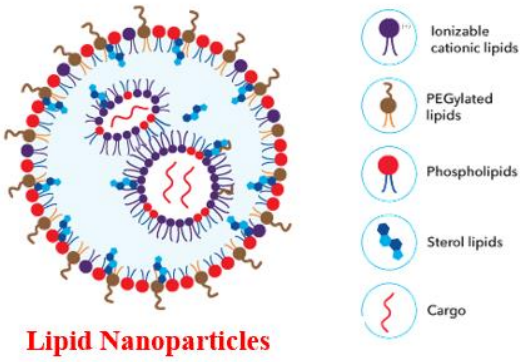


- Same size & surface properties
- Distinct rigidity

X. Ma et al *Effect of liposome rigidity on corona formation and its implications for uptake by cells.* ***In press 2026***

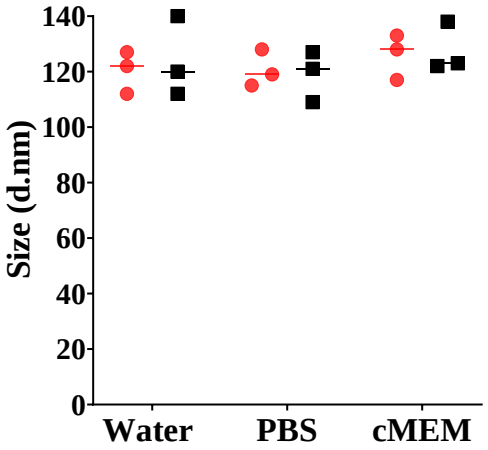
….another example: LNP and transfection efficiency

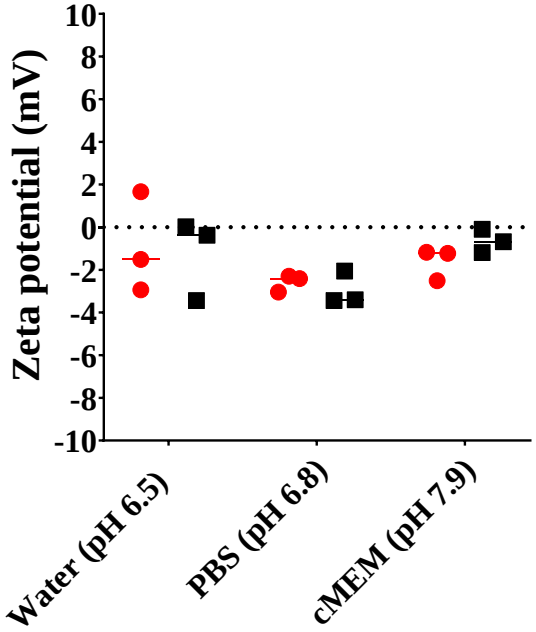
Heba Fayyaz

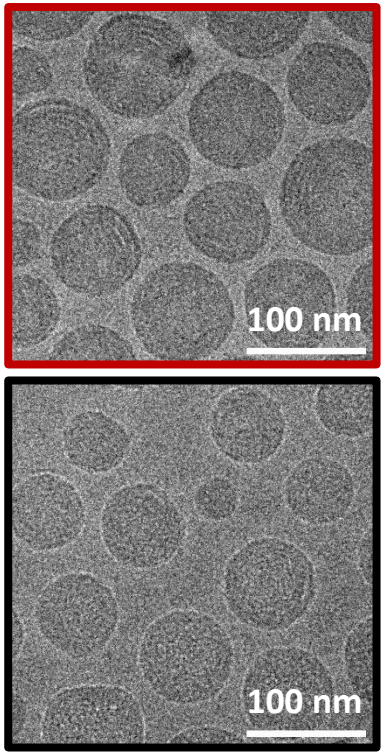


Molar ratio (Percent)				
DLin-MC3-DMA	DSPC	Cholesterol	DMG-PEG-2000	DiI
50	9.9	38.5	1.5	0.1

Poly-A loaded LNPs vs mRNA loaded LNPs







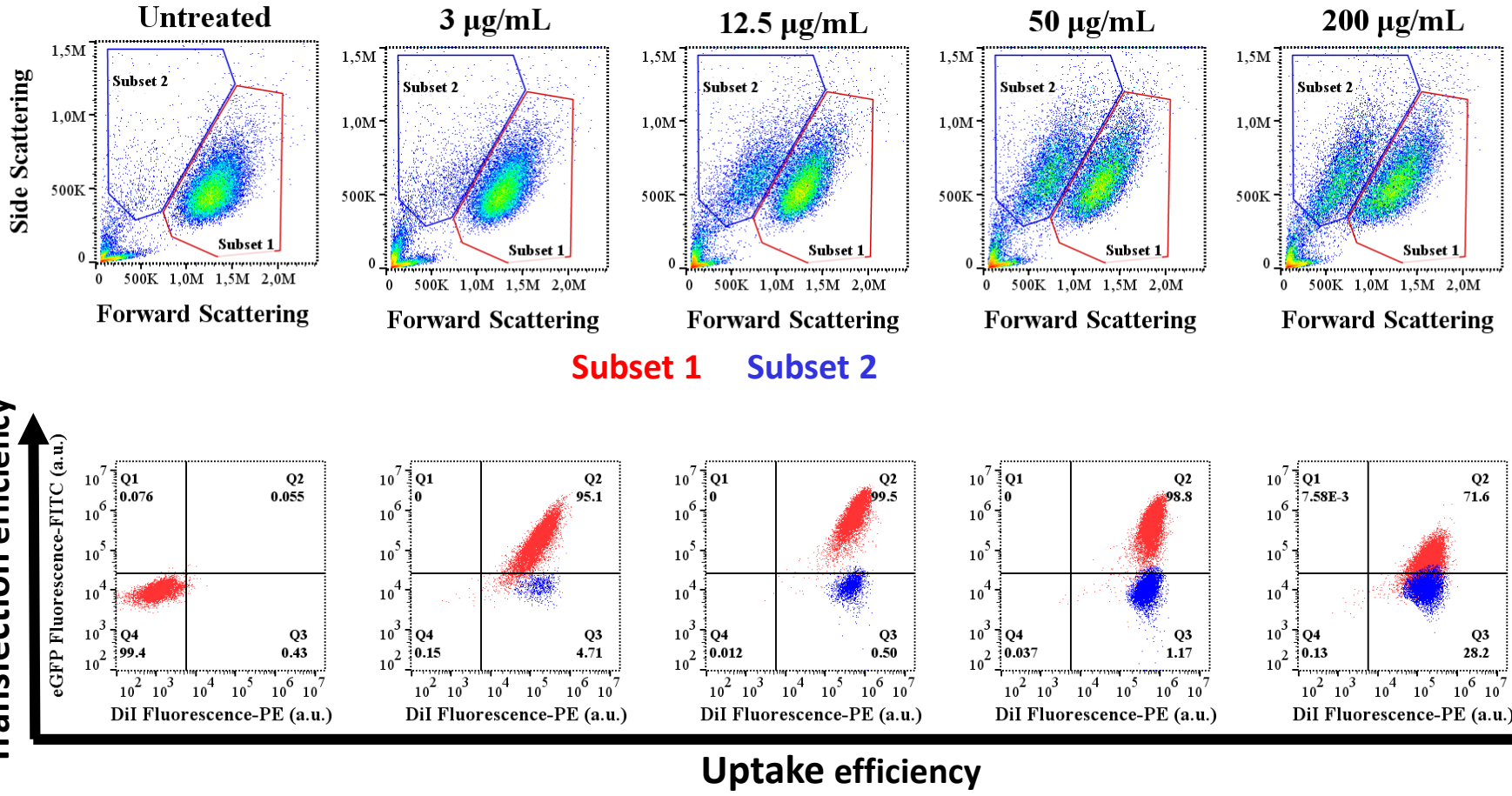
Fayyaz et al., **Materials Today Bio**, 2026, 39, 103340
Identification of a cell subpopulation with different response to lipid nanoparticles and effect of protein corona on uptake and transfection

Identification of a cell subpopulation with different response to LNPs

Heba Fayyaz

Heterogeneous cellular responses!

A second subpopulation upon incubation with LNPs!

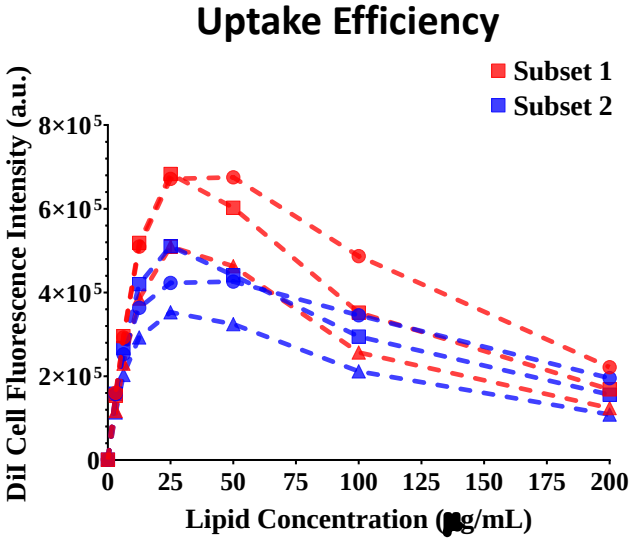


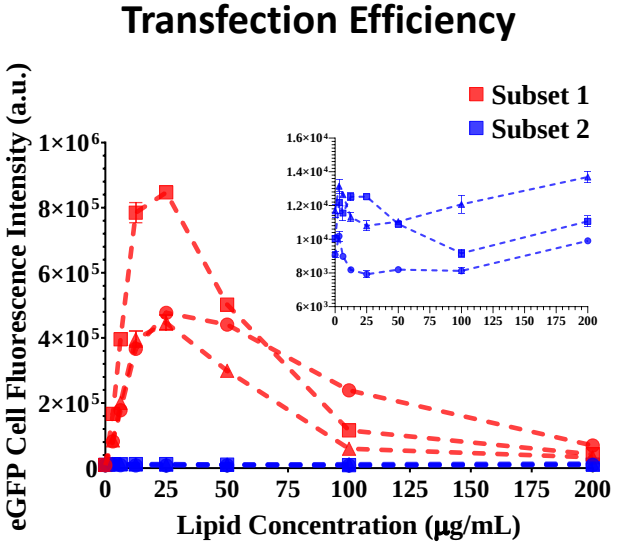
Fayyaz et al., **Materials Today Bio**, 2026, 39, 103340

Identification of a cell subpopulation with different response to lipid nanoparticles and effect of protein corona on uptake and transfection

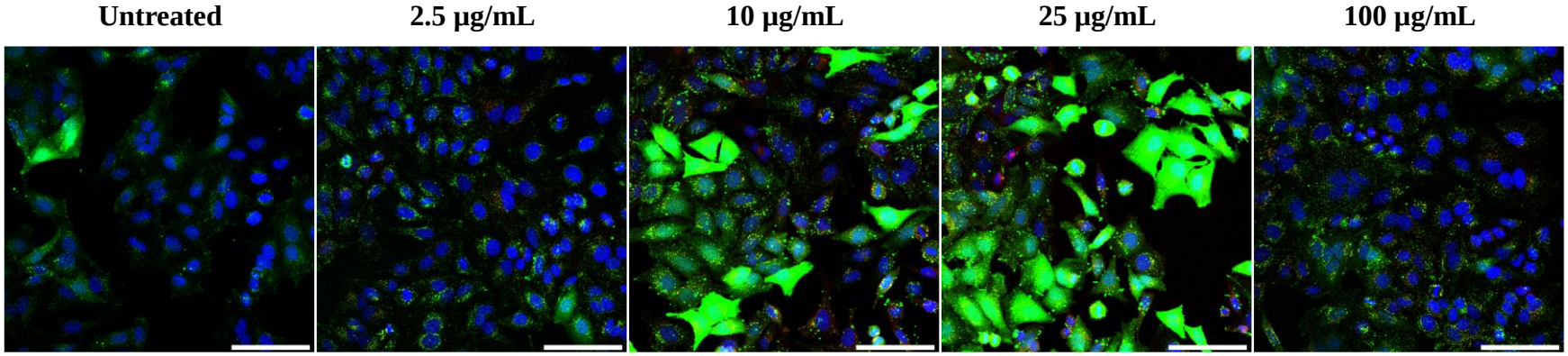
Non-monotonic dose response behaviour!

Heba Fayyaz





Endosomal Leakiness

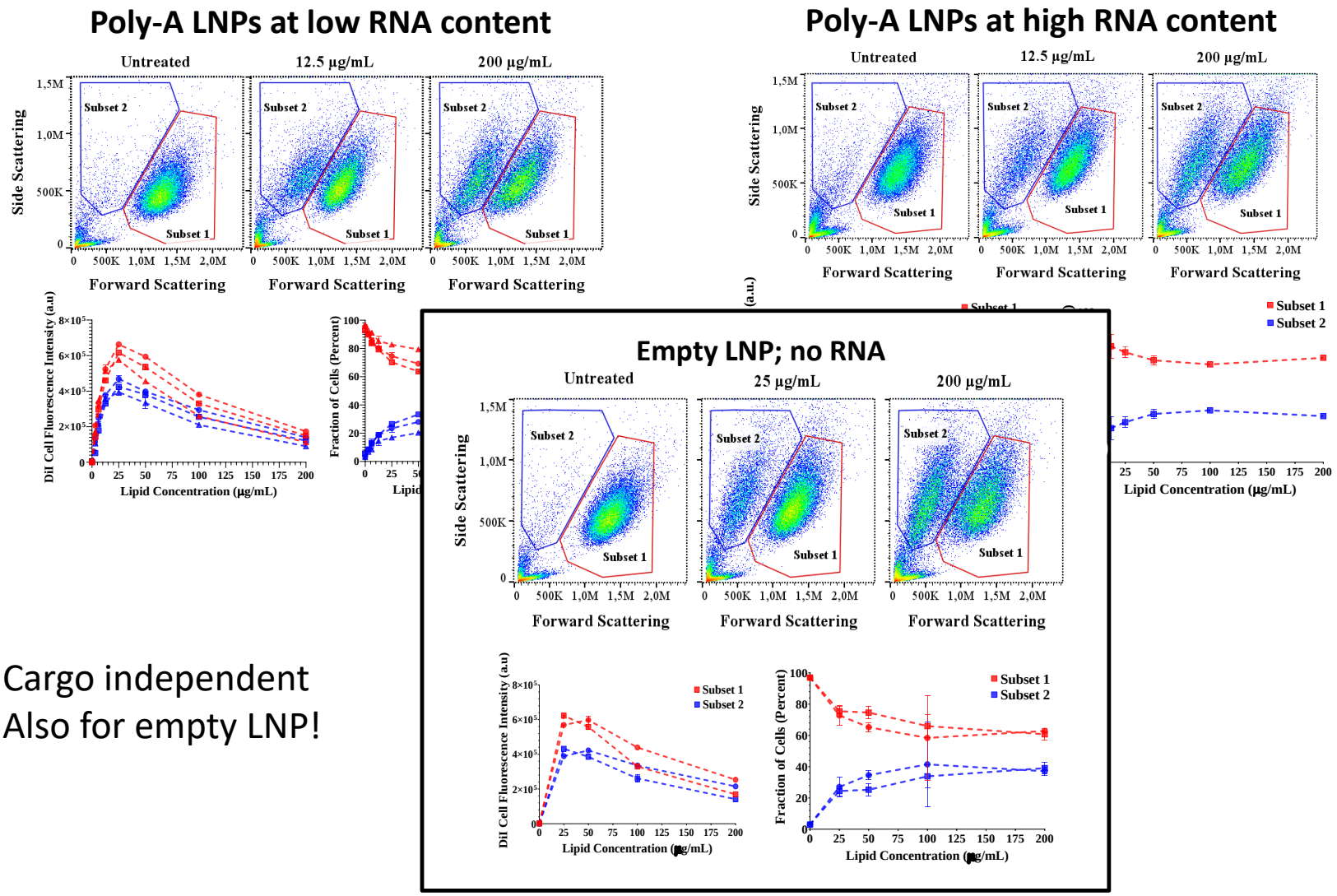


Fayyaz et al., **Materials Today Bio**, 2026, 39, 103340

Identification of a cell subpopulation with different response to lipid nanoparticles and effect of protein corona on uptake and transfection

Independent of cargo

Heba Fayyaz



- Cargo independent
- Also for empty LNP!

Fayyaz et al., **Materials Today Bio**, 2026, 39, 103340
Identification of a cell subpopulation with different response to lipid nanoparticles and effect of protein corona on uptake and transfection

…and with different ionizable lipids

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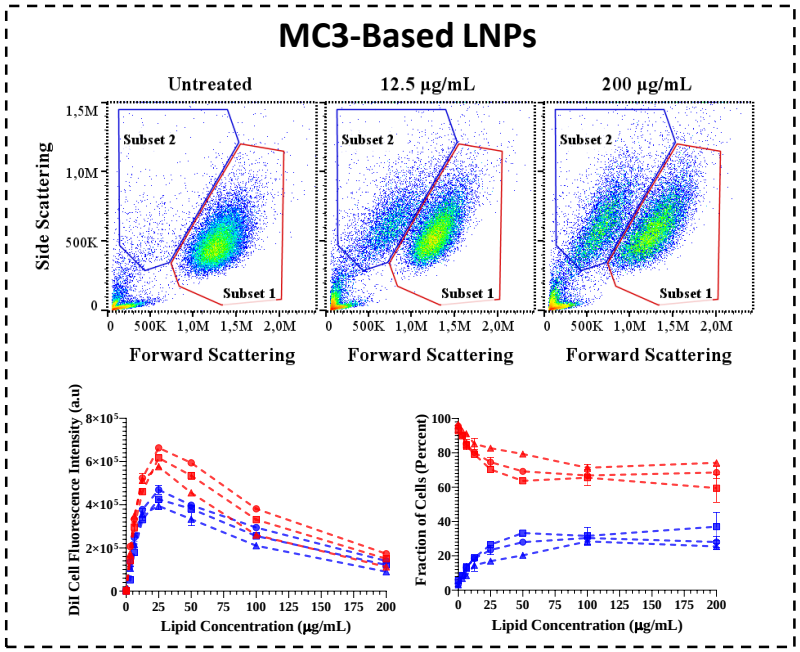

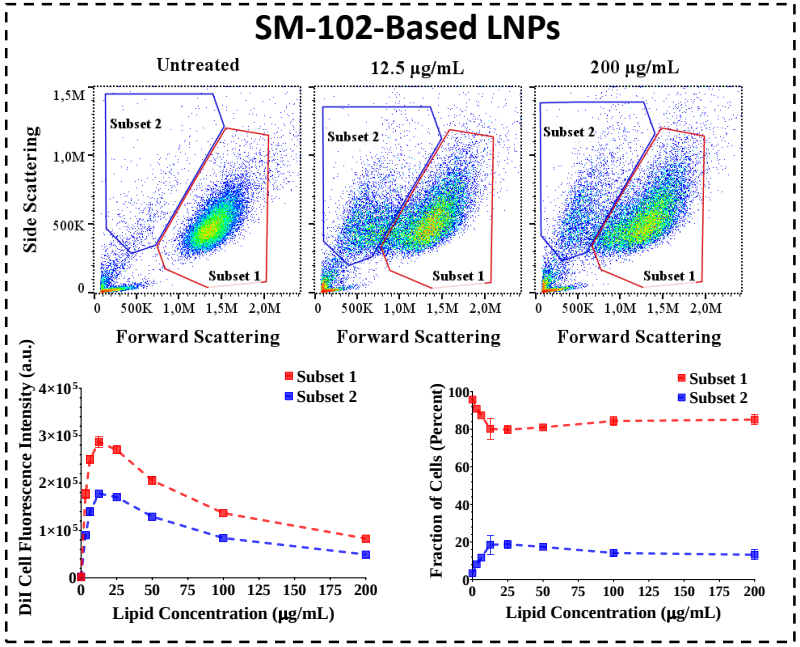

- Both with LNPs with MC3 and LNPs with SM-102

Fayyaz et al., **Materials Today Bio**, 2026, 39, 103340
Identification of a cell subpopulation with different response to lipid nanoparticles and effect of protein corona on uptake and transfection

...and in different cell types

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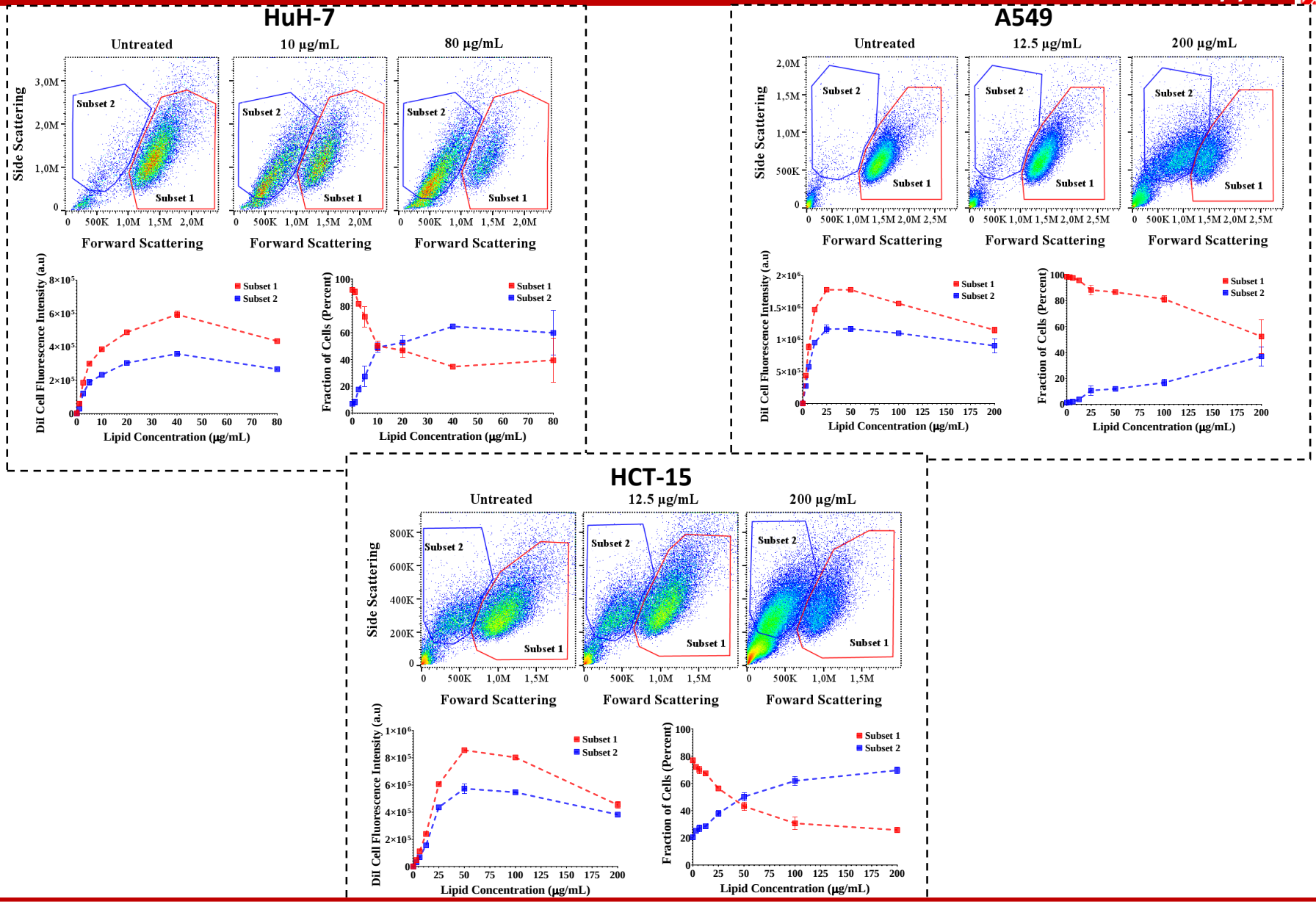

Fayyaz et al., **Materials Today Bio**, 2026, 39, 103340
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What is the second cell sub-population?

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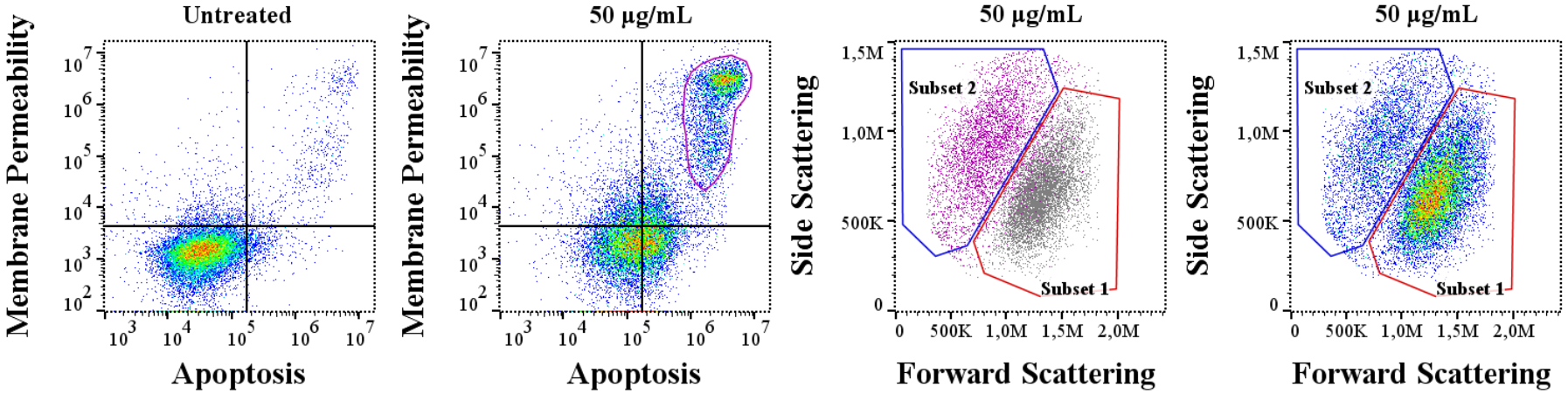
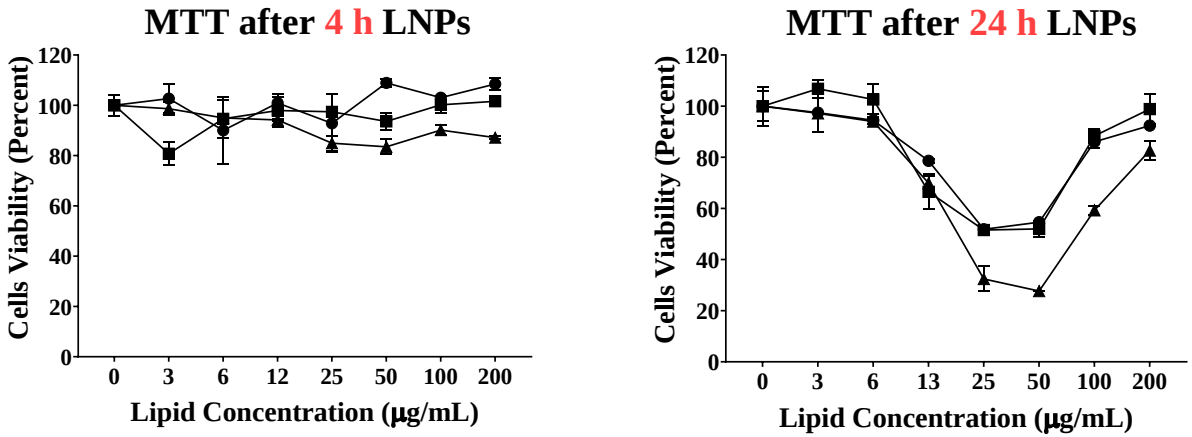
Subset 2 cells are PI- positive: a sub- population more sensitive to LNPs!



Cell viability correlated to uptake, not concentration



Fayyaz et al., **Materials Today Bio**, 2026, 39, 103340
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Why the non-monotonic uptake behavior?

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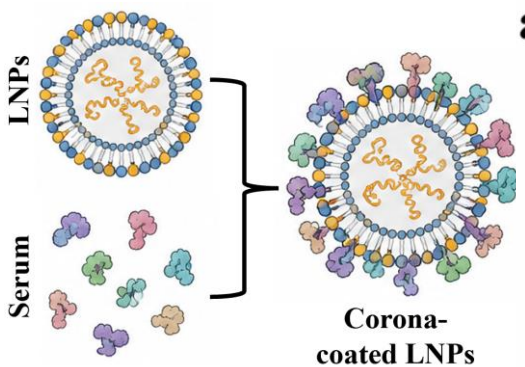
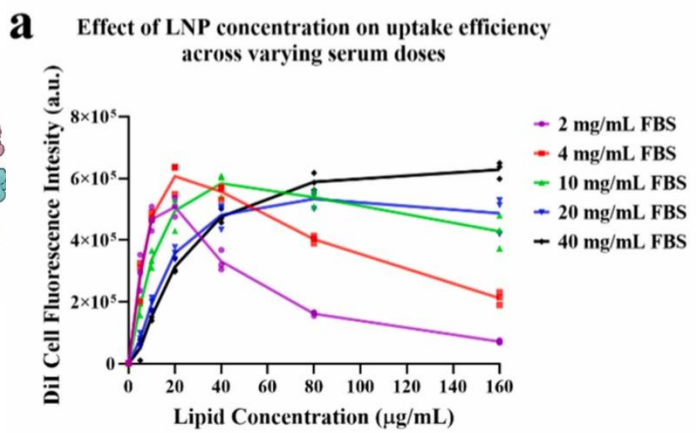
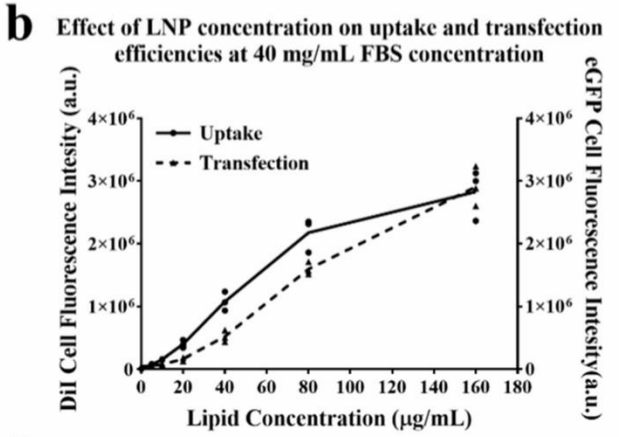
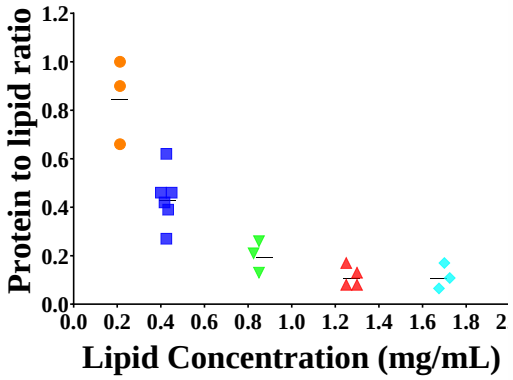
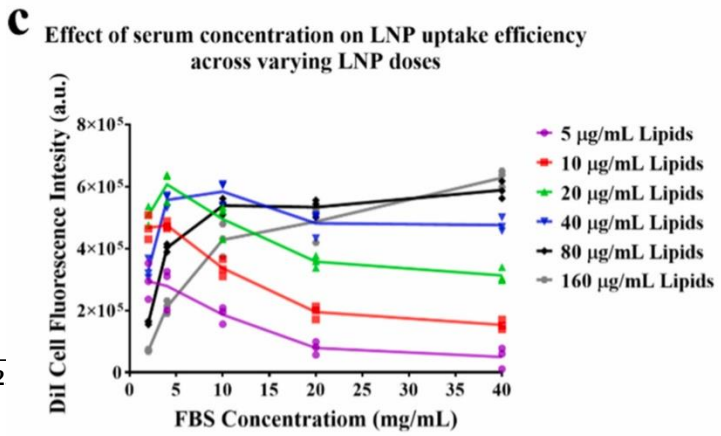
Adsorbed corona changes when changing LNP concentration

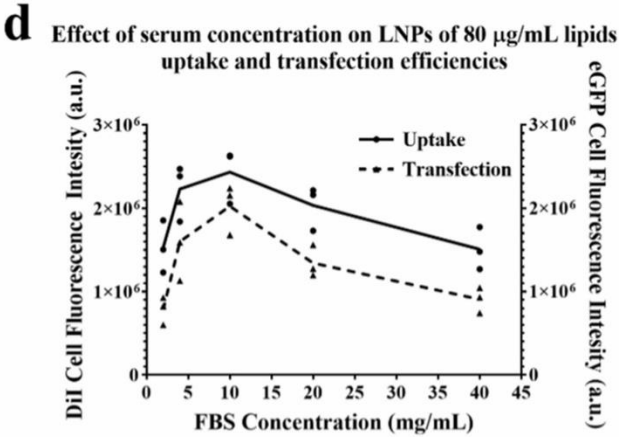












→ ***Uptake & transfection strongly modulated by exposure conditions (serum content)***

Fayyaz et al., **Materials Today Bio**, 2026, 39, 103340
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Some conclusions

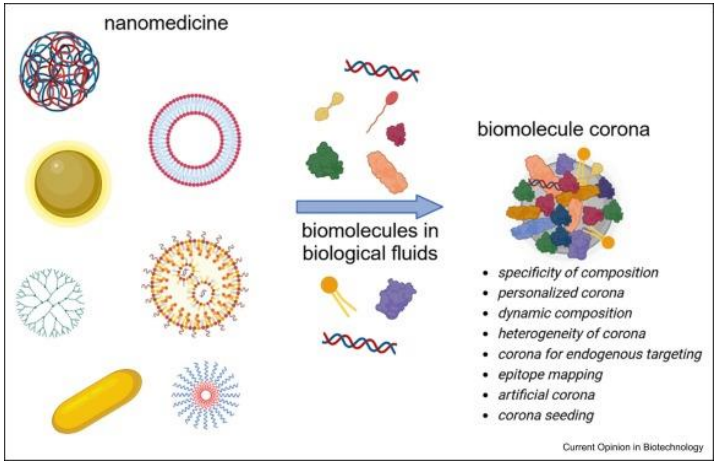
- **Why do positive nanoparticles usually have higher uptake than negative ones?**
- **Effect of liposome rigidity on corona formation and its implications for uptake by cells**
- **Effect of protein corona on LNP uptake and transfection**

→ ***Nanomedicine interactions with biological fluids shape their cell behavior***

Salvati, ***Current Opinion Biotech****, 2024,* 87, 103101

The biomolecular corona of nanomedicines: effects on nanomedicine outcomes and emerging opportunities

Special Issue ***Nanobiotechnology (2024)*** Edited by Warren C.W. Chan University of Toronto, Canada



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