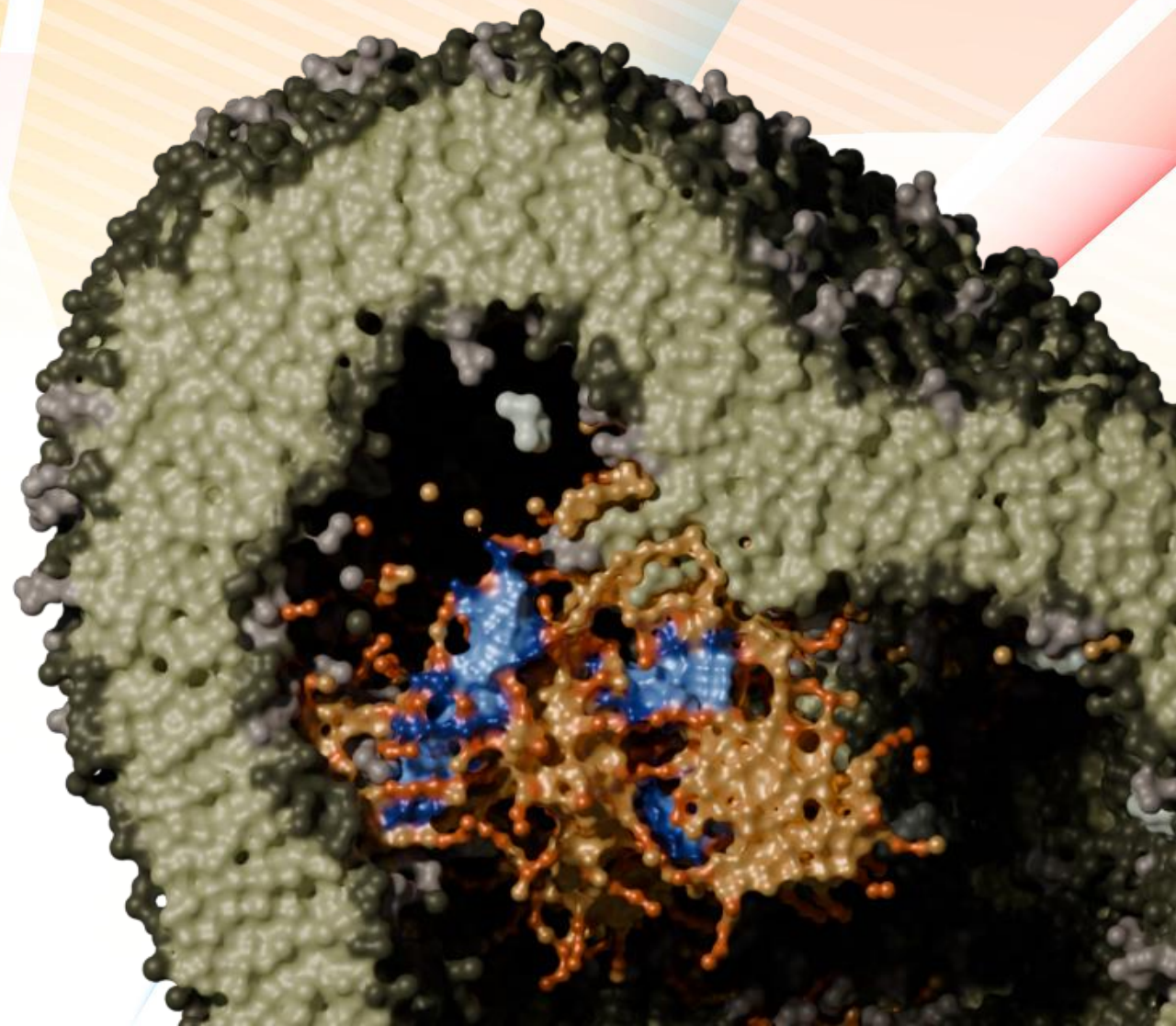


proton sponge or membrane fusion? – endosomal escape of siRNA polyplexes unveiled by MD simulations

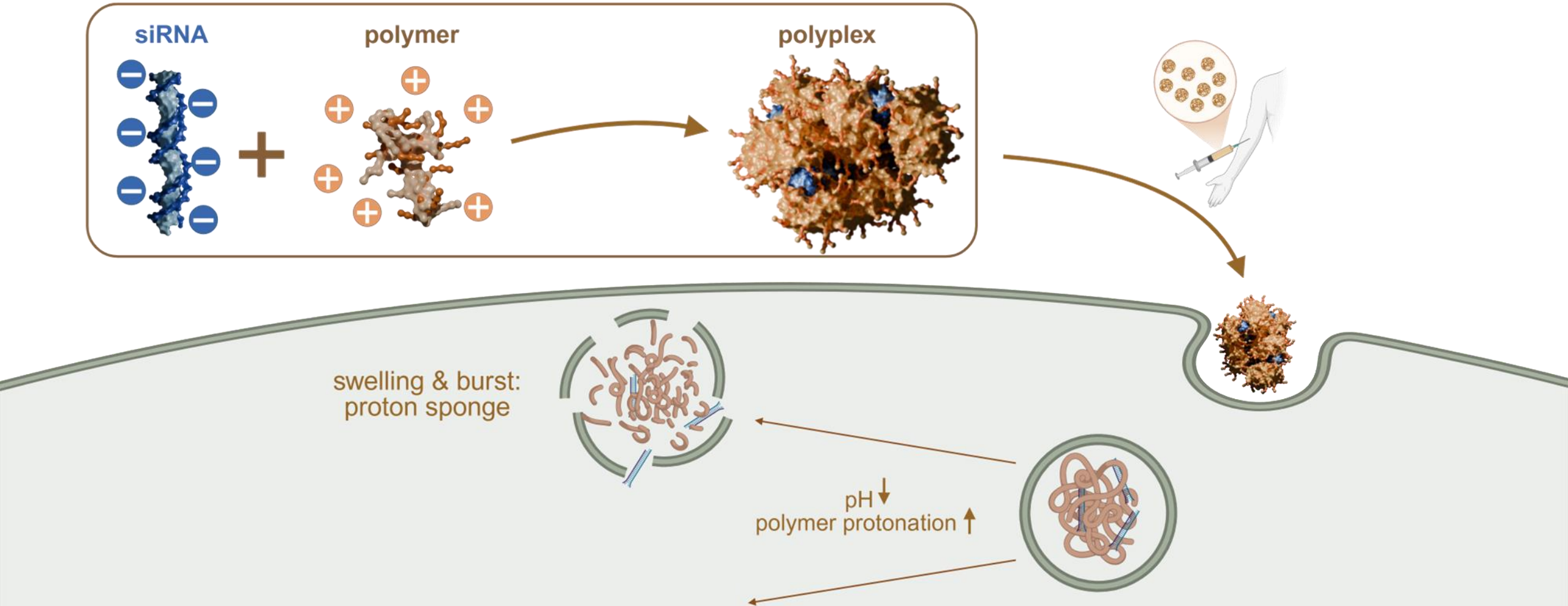
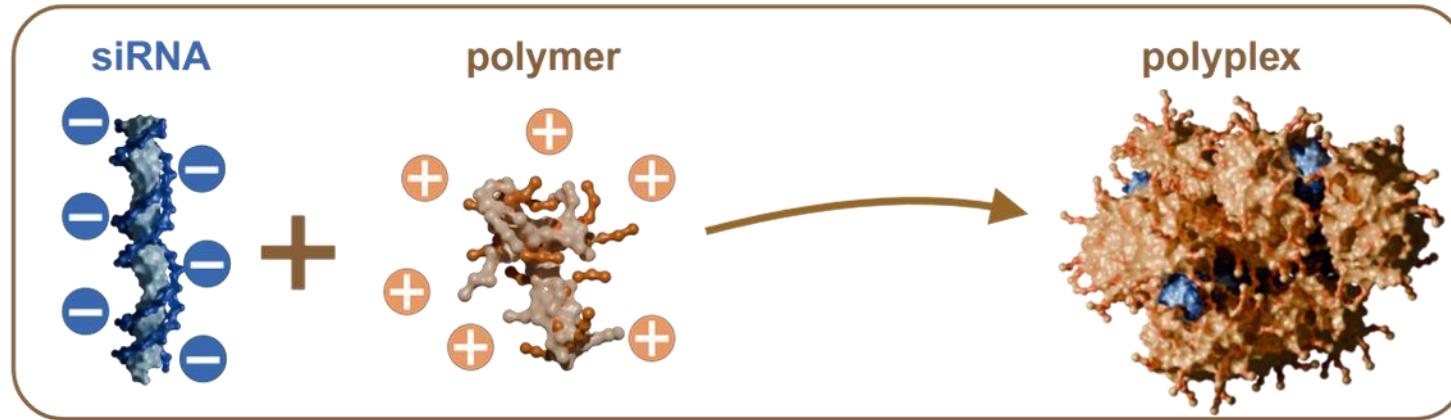
Katharina M. Steinegger

group of Prof. Olivia M. Merkel

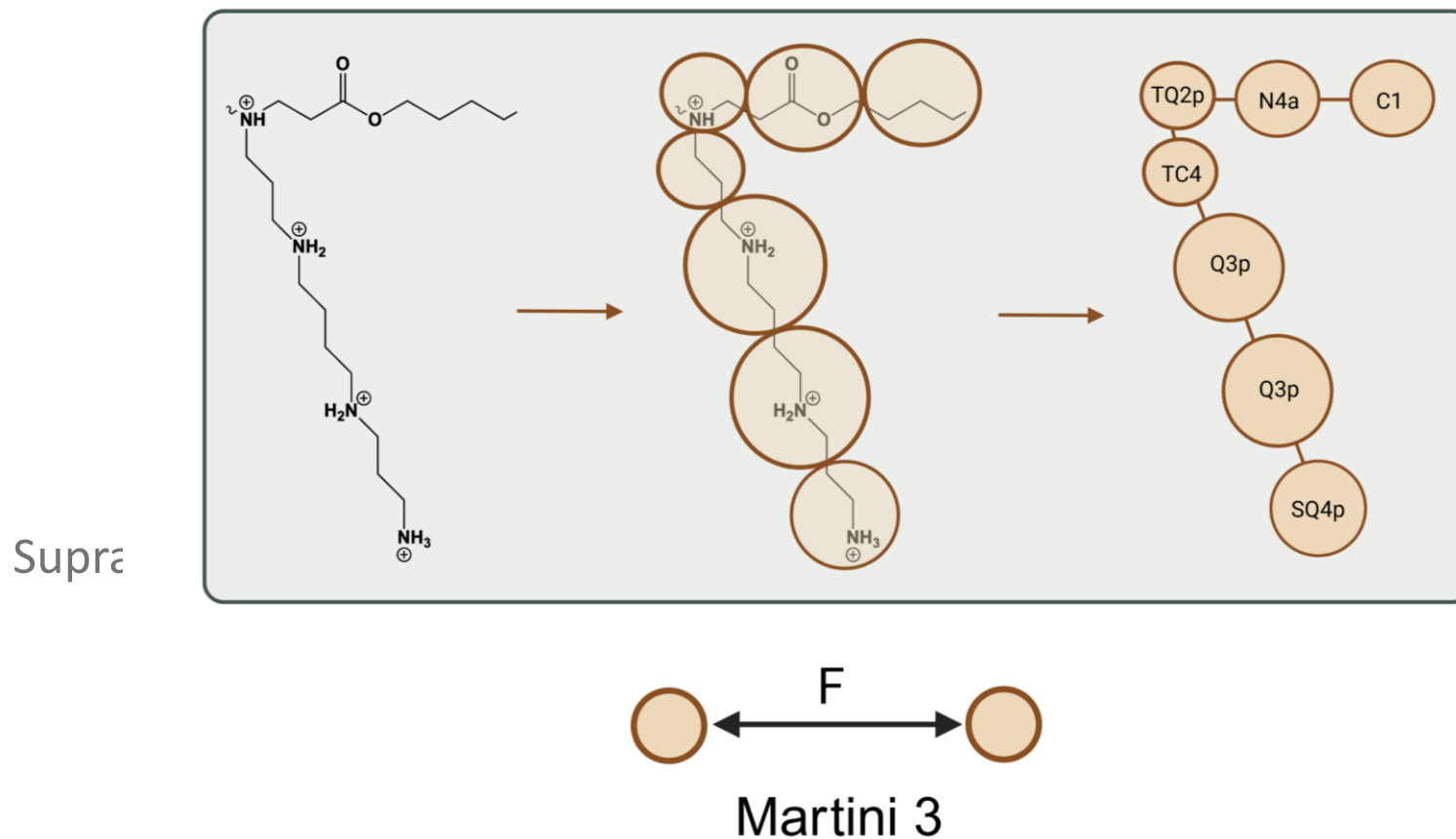
LMU Munich



siRNA technology and the question of endosomal escape



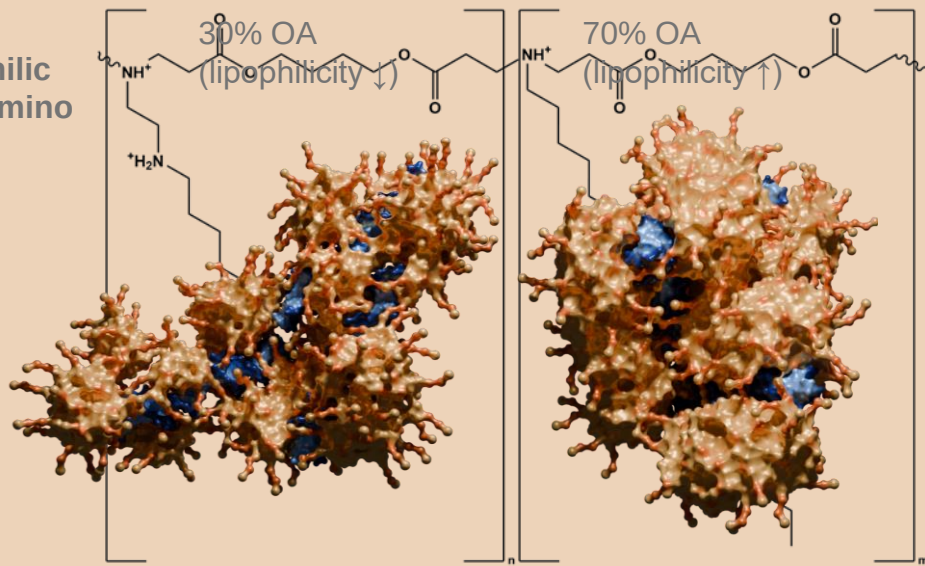
Coarse Grained (CG) Molecular Dynamics simulations



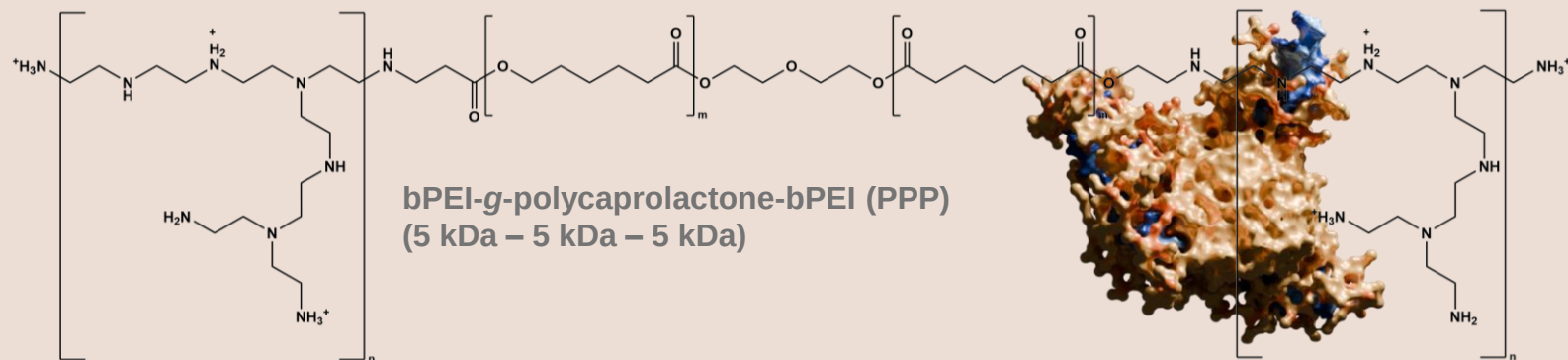
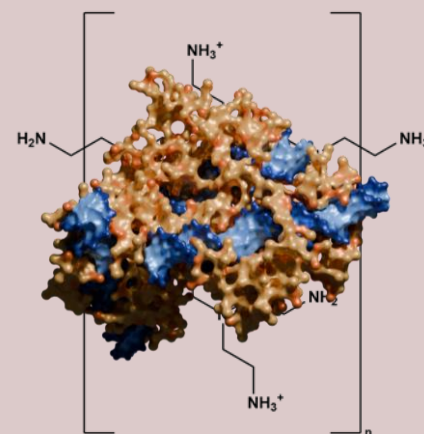
Adapted from Ingolfsson et al. Computational 'microscopy' of cellular membranes, Journal of Cell Science (2016) 129, 257-268 doi:10.1242/jcs.176040

particle formulations

9 kDa
amphiphilic
poly(β -amino
ester)
(PBAE)

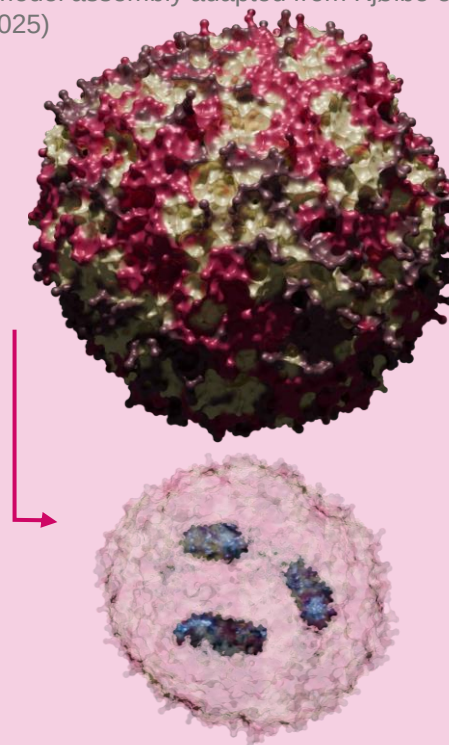


25 kDa branched polyethyleneimine
(bPEI)



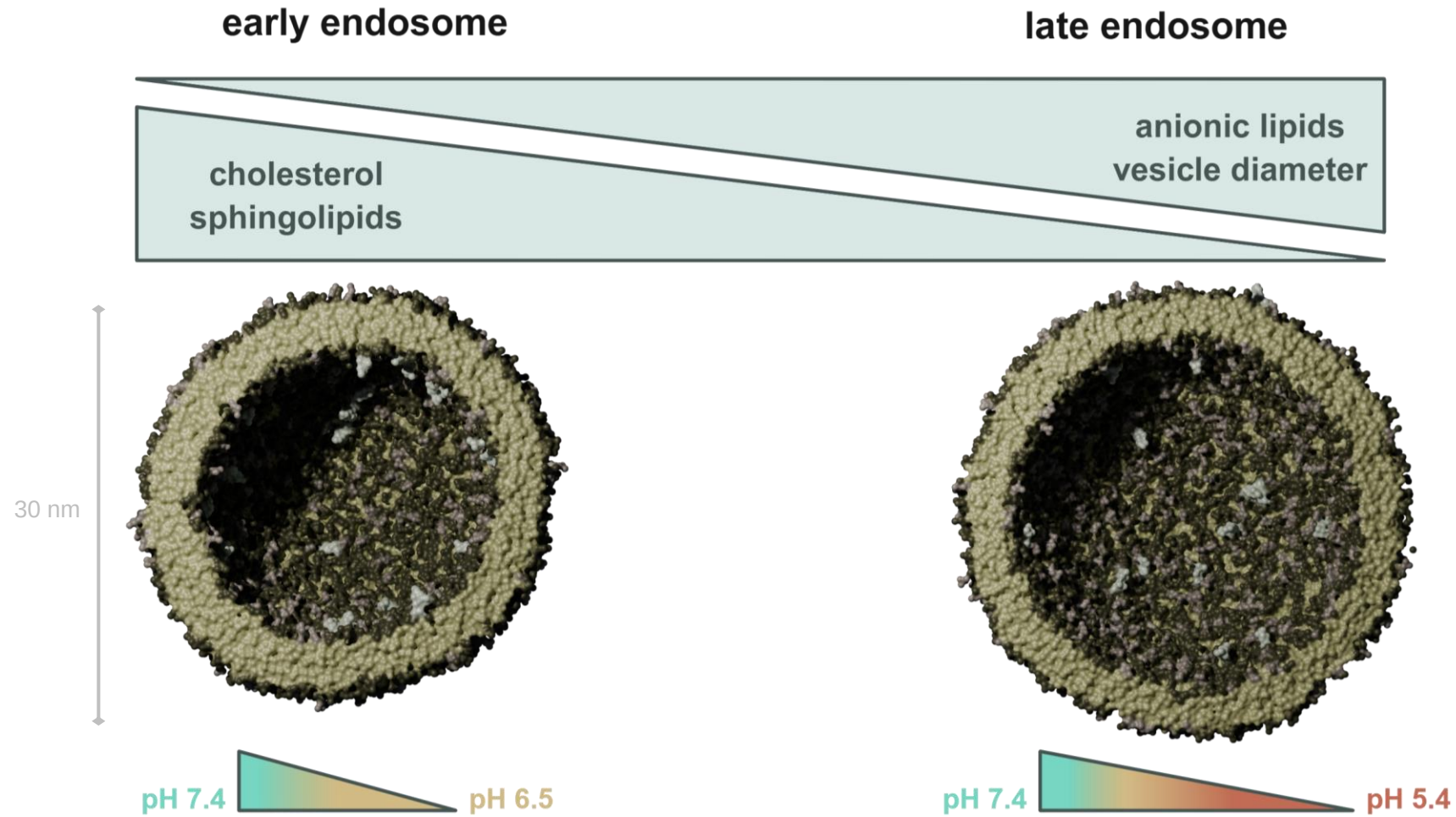
Onpattro® - like LNP

(model assembly adapted from Kjølbe et al. 2025)



16 nm

model membranes & vesicles



proton sponge or membrane fusion?

70% OA PBAE

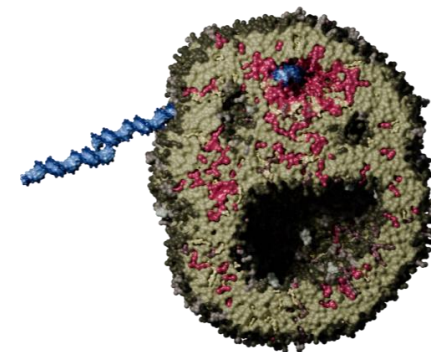
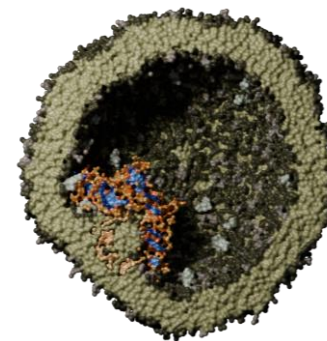
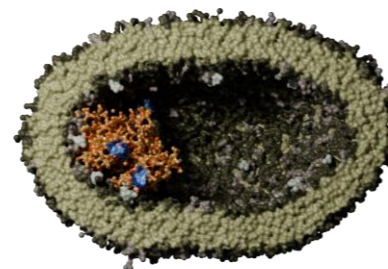
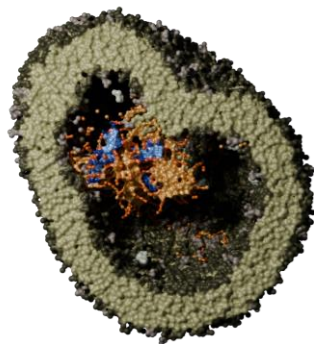
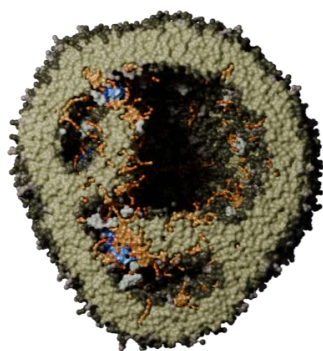
30% OA PBAE

bPEI

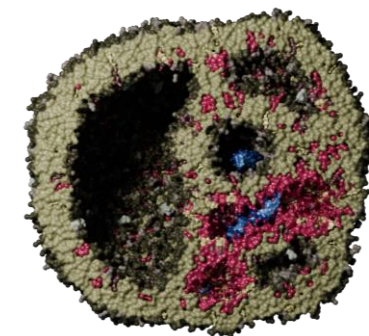
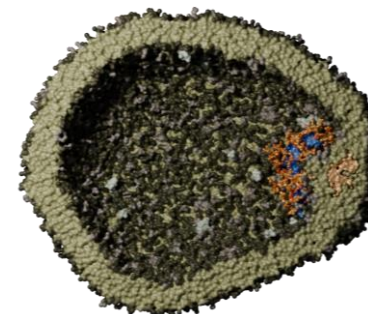
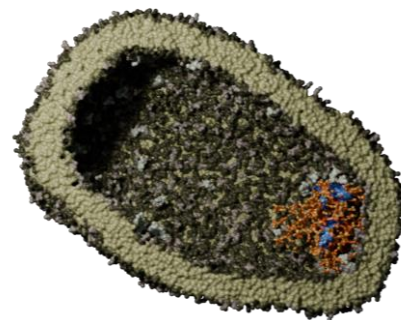
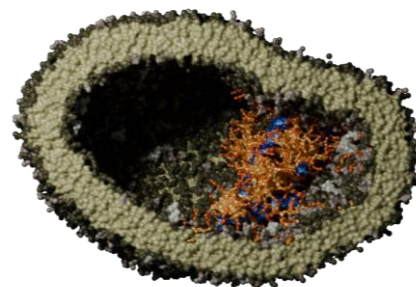
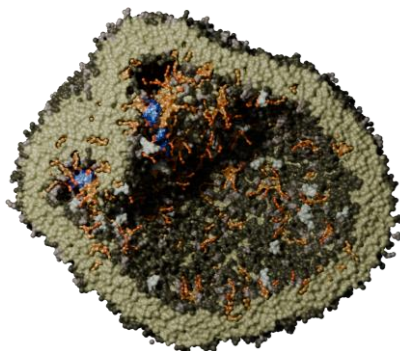
PPP

LNP

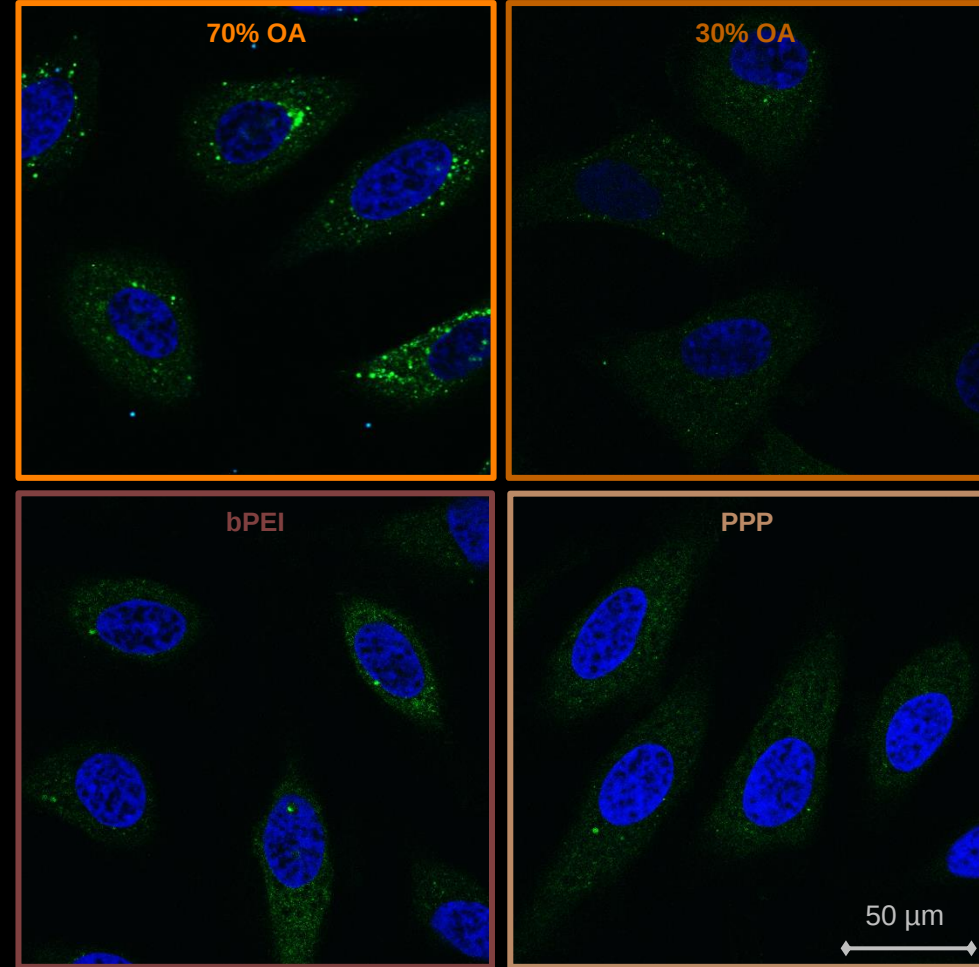
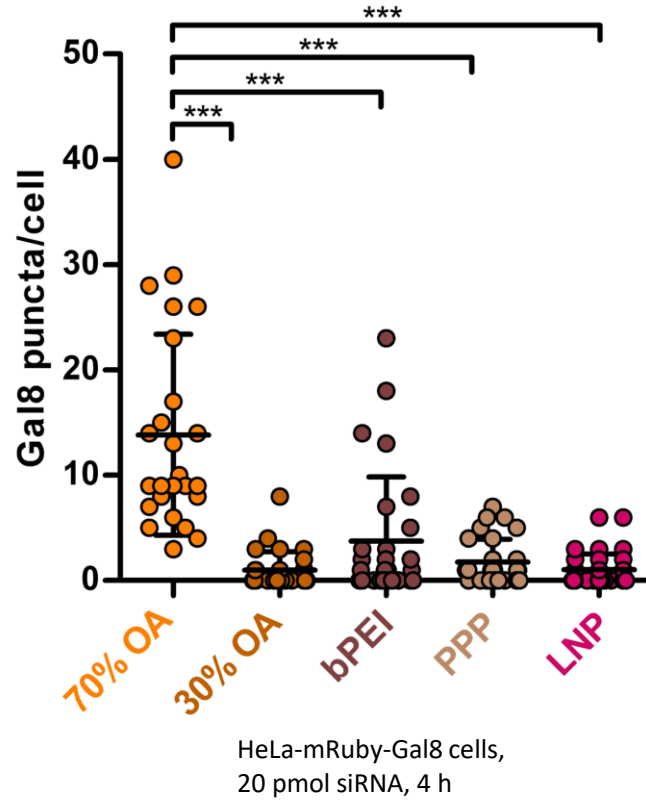
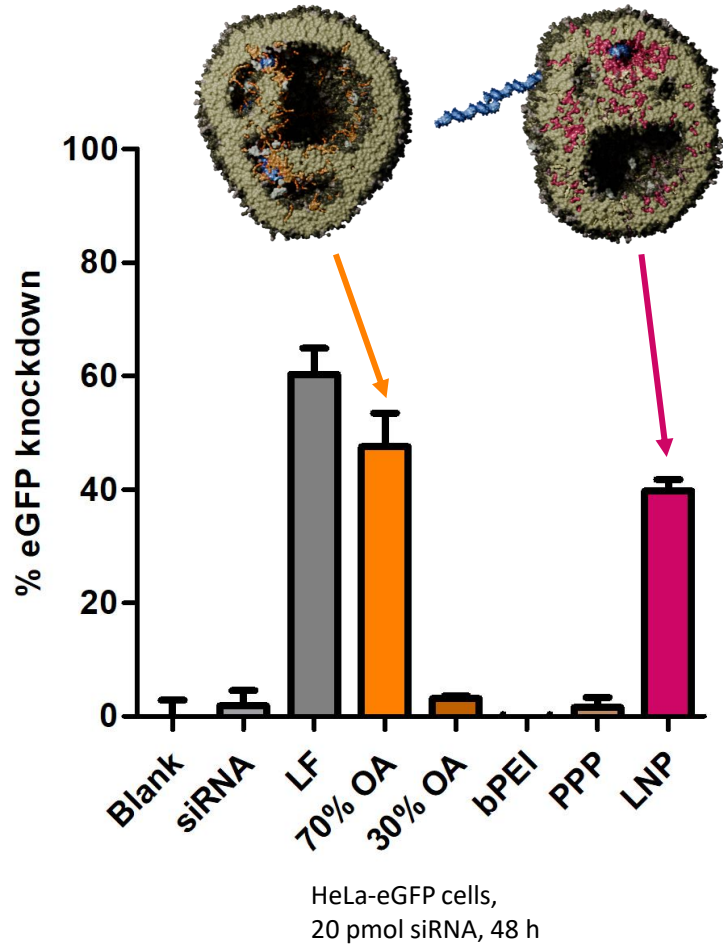
Early
Endosome



Late
Endosome



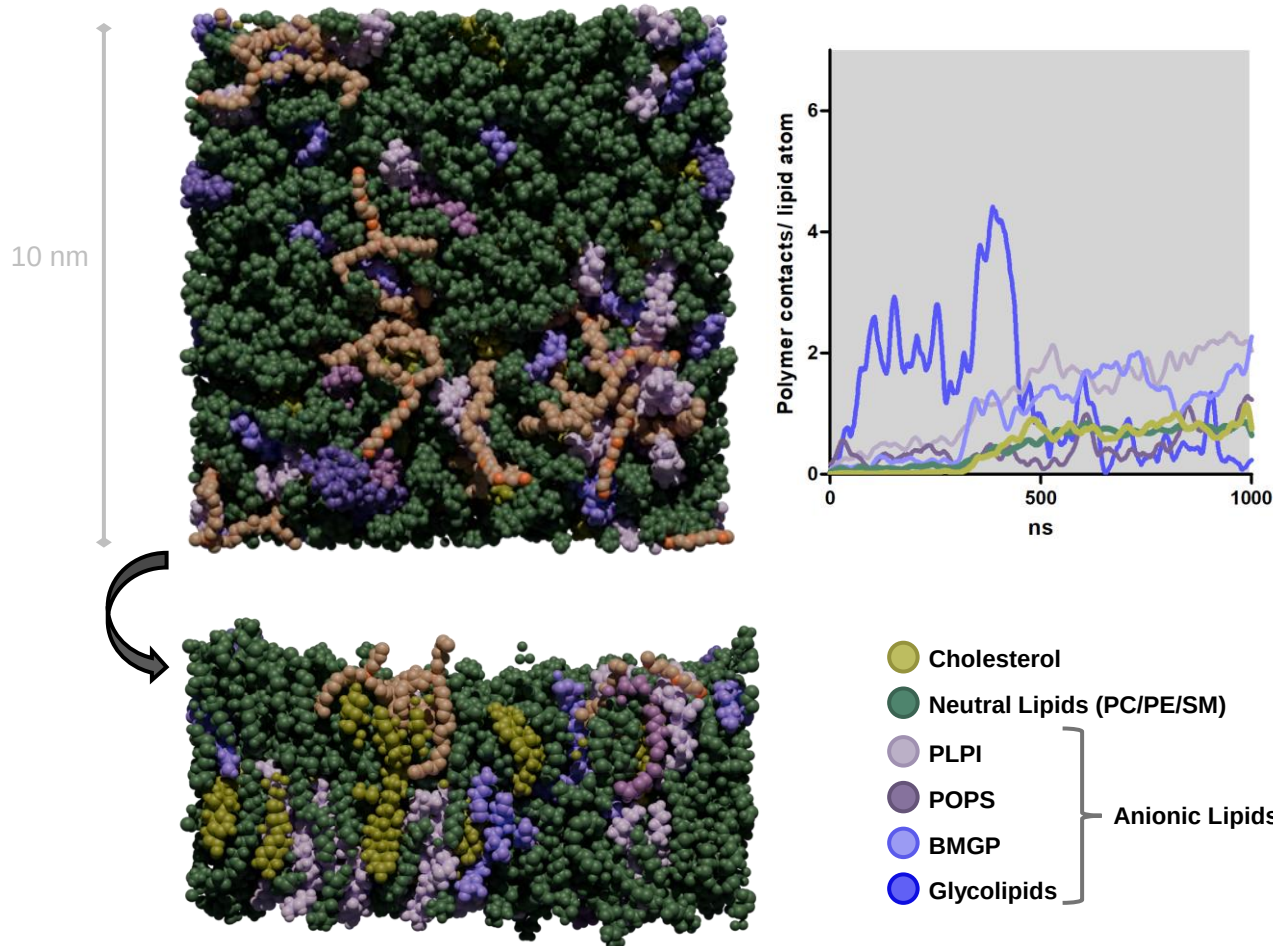
in vitro performance in HeLa cells



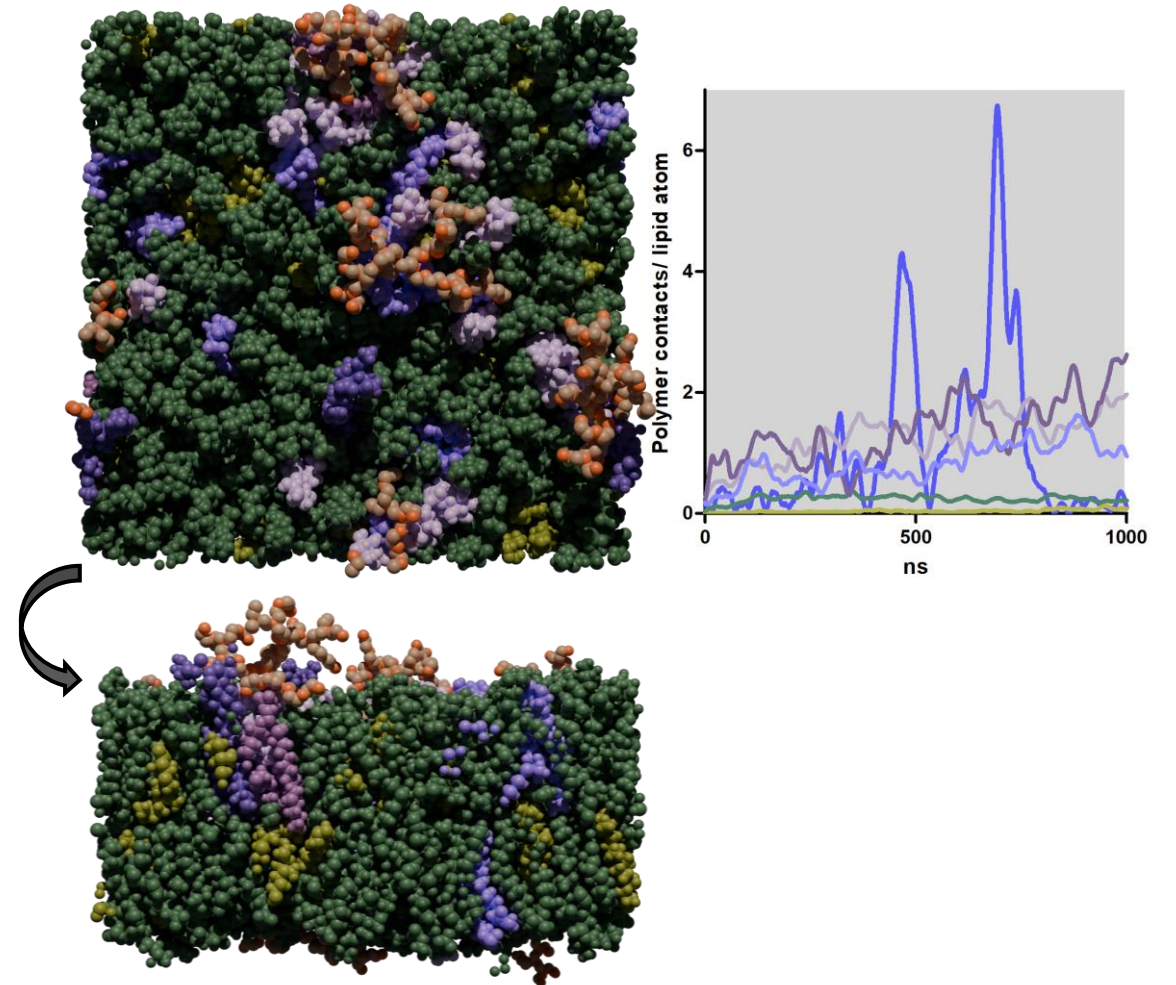
the role of anionic lipids

All Atom simulations: polymer + late endosomal membrane

70% OA PBAE

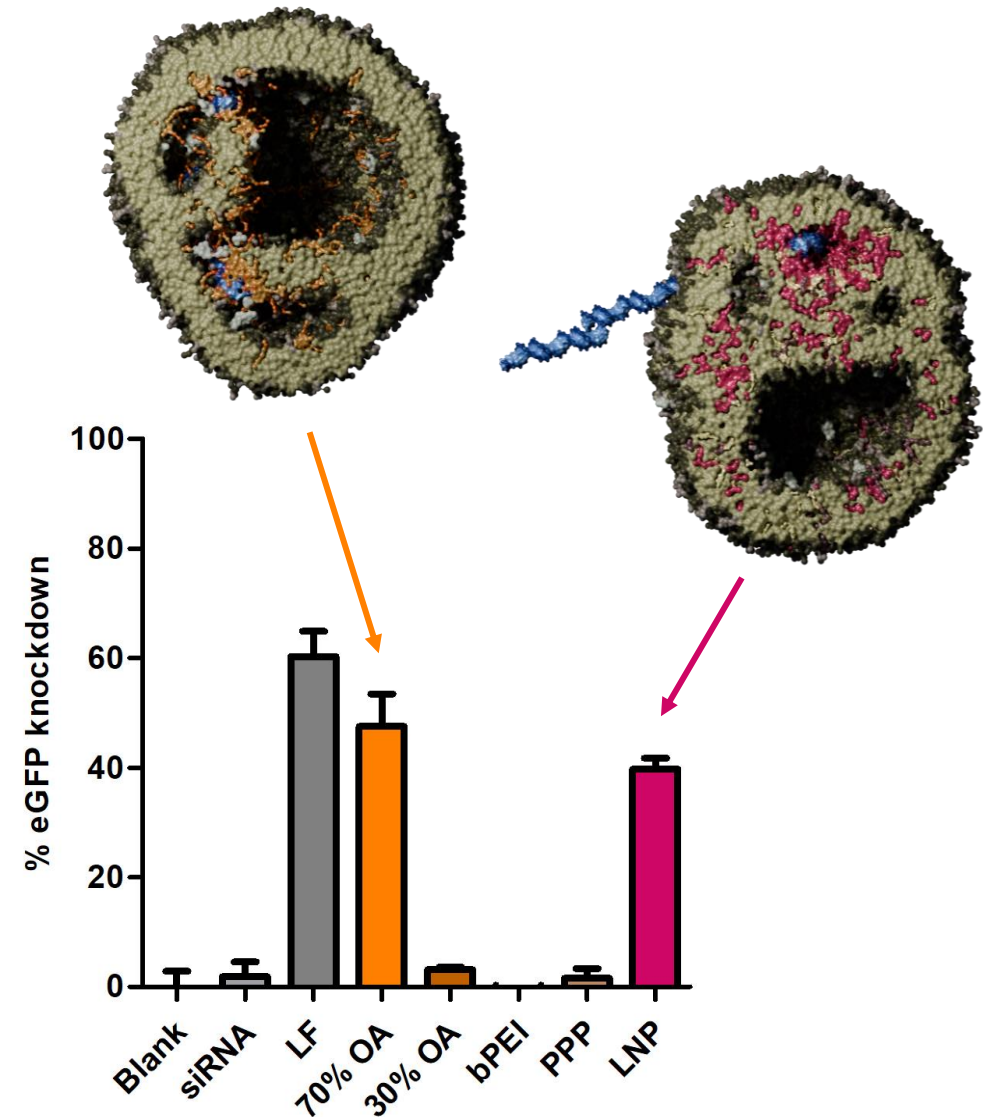


bPEI



conclusion

- Hydrophobic interactions increase endosomal escape (EE) efficiency
 - Membrane disturbance
 - Particle disintegration
- Not one mechanism applies to the EE of all polyplexes
- Anionic lipids in endosomal membranes cluster around cationic nanoparticle materials (polymer/ ionizable lipid)





LUDWIG-
MAXIMILIANS-
UNIVERSITÄT
MÜNCHEN



Thank you!

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Dr. Benjamin Winkeljann

Min Jiang



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PHILADELPHIA, PA
JULY 13-18, 2025

**NEXT-GENERATION
DELIVERY INNOVATIONS**