Replacing PEG-Lipid with Amphiphilic Polycarbonates in mRNA-Loaded Lipid Nanoparticles

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Introduction

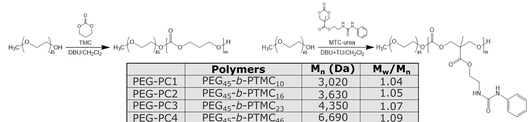
• PEG-lipid is a critical component of lipid nanoparticles (LNPs) which assists in preventing particle aggregation in blood and avoiding sequestration by the mononuclear phagocyte system, prolonging their systemic circulation.[1,2]

• Herein, we synthesized a series of polycarbonate-lipid hybrid mRNA nanoparticles (PC-HNPs) by substituting amphiphilic PEG-polycarbonate diblock copolymers for ALC-0159.[3,4]

Highlights

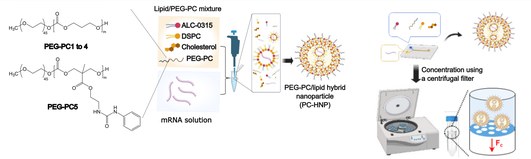
◆ Variation of PC block lengths significantly influenced mRNA encapsulation efficiency, colloidal stability and PEG shedding of PC-HNPs.
◆ PC3- and PC4-HNPs had higher mRNA translation and endosomal escape efficiency than ALC-LNP formulation.
◆ Upon s.c. injection, PC3-HNP achieved comparable lymph node accumulation to ALC-LNP while avoiding undesirable liver accumulation.

PEG-polycarbonate (PEG-PC)

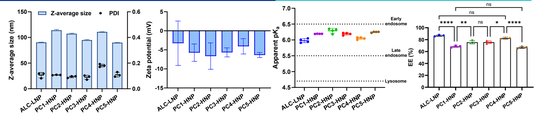


	Polymers	M_n (Da)	M_w/M_n
PEG-PC1	PEG_{45}-*b*-$PTMC_{10}$	3,020	1.04
PEG-PC2	PEG_{45}-*b*-$PTMC_{16}$	3,630	1.05
PEG-PC3	PEG_{45}-*b*-$PTMC_{23}$	4,350	1.07
PEG-PC4	PEG_{45}-*b*-$PTMC_{46}$	6,690	1.09
PEG-PC5	PEG_{45}-*b*-P(MTC-urea)$_4$	3,290	1.11

Methodology

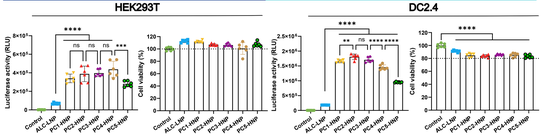


EE increased with longer PTMC length

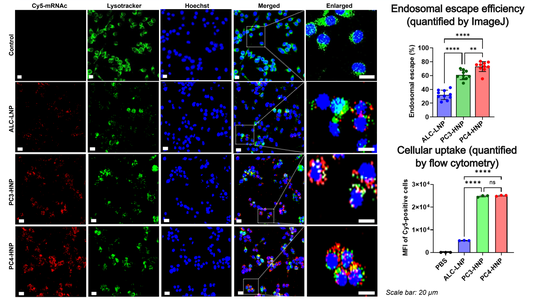


• EE gradually increased with increasing DP of PTMC block from 10 to 46, suggesting that increasing hycrophobicity of longer PTMC blocks rendered more stable mRNA encapsulation.

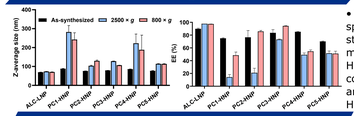
PC-HNPs offered significantly higher FLuc mRNA translation efficiency



PC3- & PC4-HNP escaped endosomes more efficiently

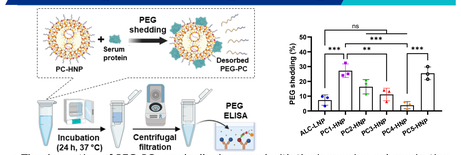


PC3-HNP showed the highest colloidal stability



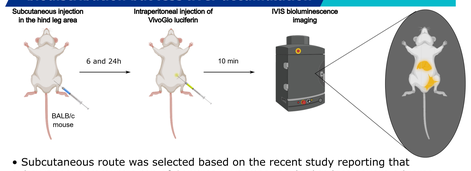
• The centrifugation speed affected the structure and EE of most PC-HNPs. PC3-HNP had the best colloidal stability among the tested PC-HNPs.

PEG shedding reduced with longer PTMC length

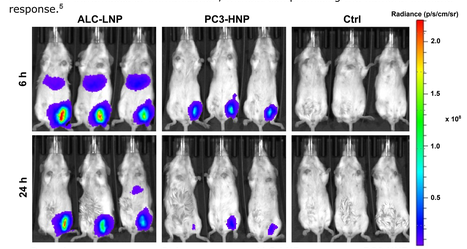


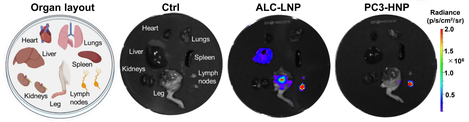
• The desorption of PEG-PCs gradually decreased with the increasing polymerization degree of PTMC blocks. The longer PTMC blocks are thought to exhibit stronger hydrophobic interactions with adjacent lipid molecules, thus resulting in a tighter anchoring of PEG chains on the particle surface.

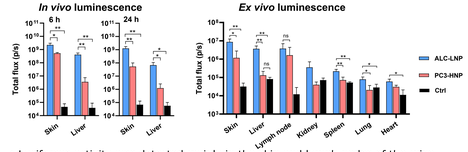
PC3-HNP exhibited comparable lymph node biodistribution but less liver accumulation



• Subcutaneous route was selected based on the recent study reporting that subcutaneous immunization of Comirnaty vaccine resulted in less severe adverse events than intramuscular immunization, without compromising the humoral immune response.[5]







• Luciferase activity was detected mainly in the skin and lymph nodes of the mice treated with PC3-HNPs. Of note, PC3-HNP achieved comparable lymph node accumulation to ALC-LNP while avoiding liver accumulation.

References

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Acknowledgements

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