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CRS Astrocyte Membrane-Coated Nanoparticles: A Targeted Therapeutic Strategy for Glioblastoma Treatment

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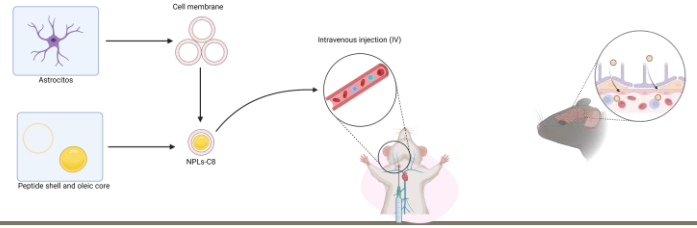
Background

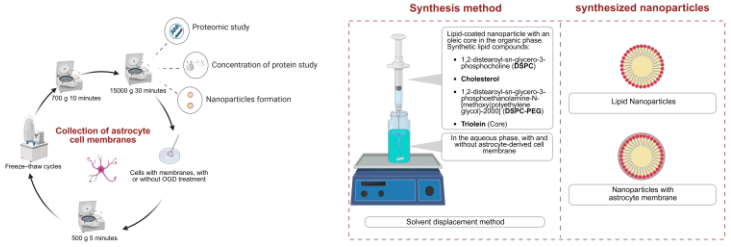
The **blood-brain barrier** (BBB) is one of the major challenges in the treatment of glioblastoma (GBM), a highly aggressive brain tumor with poor response to conventional therapies (1). Due to its **low permeability**, the BBB prevents many drugs from reaching the tumor site at therapeutic concentrations, significantly limiting their clinical efficacy (2).

In this study, we developed **lipid-based nanoparticles (NPLs) coated with astrocyte membranes (NPL-C8)** to enhance **biocompatibility**, facilitate BBB crossing, and promote **selective uptake** by tumor cells.

Astrocytes are responsible **for synthesizing lipoproteins** that nourish **glioblastoma cells** (4), which exhibit a high demand for cholesterol and **overexpress low-density lipoprotein receptors (LDLR)** (5). Thus, the use of astrocyte-derived membranes may more accurately mimic endogenous al nervous system**, improving nanoparticle internalization** by GBM cells.



Methods



Results

The physicochemical properties of NPLs were characterized by DLS, TEM, and zeta potential analysis. The impact of astrocyte membrane coating on particle size, homogeneity, morphology, and surface charge was evaluated. These analyses provide insight into the structural integrity and colloidal stability of the coated nanoparticles.

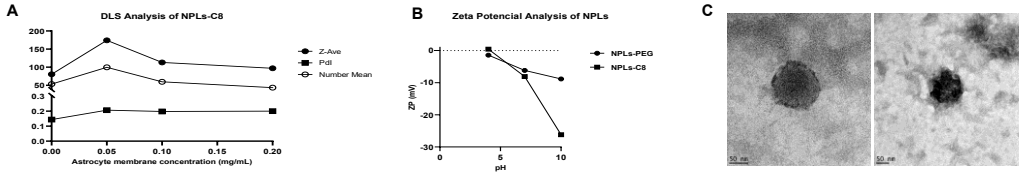


Figure 1. Physicochemical characterization of NPL formulations.(**A**) DLS analysis of NPLs formulated with different concentrations of astrocyte membrane. Particle size (Z-Ave) increased with membrane concentration until reaching a plateau. The polydispersity index (PdI) remained stable (~0.2), indicating a uniform size distribution. Particle number followed a similar trend, suggesting efficient and homogeneous NPL formation. (**B**) Zeta potential analysis of NPLs at different pH values. Formulations without membrane and with 0.2 mg/mL membrane were tested. Zeta potential decreased with increasing pH and became more negative in membrane-coated formulations, indicating that the presence of the membrane alters the surface charge profile. (**C**) Representative TEM images of NPLs (NPL-PEG and NPL-C8). Both formulations exhibited spherical morphology with diameters between 50 and 100 nm, consistent with the DLS results.

To evaluate cellular uptake of NPL formulations, in vitro assays were performed using C8-D1A, GL-261, and bEnd-3 cells. Internalization of uncoated NPLs, NPLs coated with astrocyte membrane (0.2 mg/mL), and NPLs coated with paraformaldehyde (PFA)-fixed membrane—to block membrane protein function—was compared. This allowed determination of the role of membrane proteins in the cellular uptake process.

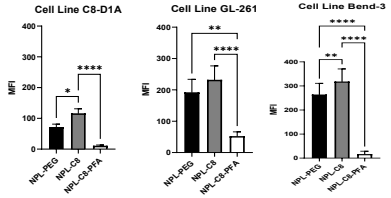


Figure 2. In vitro uptake of NPLs in neuronal and endothelial cell lines.Cellular internalization of NPLs was evaluated in C8-D1A, GL-261, and bEnd-3 cells. Three nanoparticle conditions were tested: NPLs-PEG, NPLs-C8, and NPLs coated with paraformaldehyde (PFA)-fixed membranes. A significant reduction in uptake was observed in all cell lines with PFA-fixed membrane-coated NPLs, highlighting the role of membrane proteins in nanoparticle internalization. Data are presented as mean ± SD (N = 3).

Pharmacokinetic profiles of membrane-coated and uncoated NPLs were analyzed after systemic administration. Monitoring their presence in blood over time allowed evaluation of the effect of membrane coating on circulation stability and clearance rate.

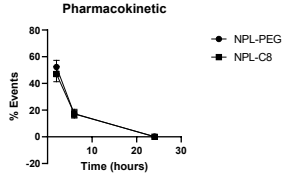


Figure 3. Pharmacokinetic profiles of membrane-coated and uncoated NPLs after systemic administration. Both membrane-coated and uncoated NPL formulations showed similar blood circulation kinetics. Data are presented as percentage of events ± SD (n = 3).

To assess the ability of NPLs to cross the blood–brain barrier (BBB), brain distribution was analyzed 24 h after systemic injection using IVIS imaging. Membrane-coated and uncoated formulations were compared to evaluate the impact of membrane coating on brain delivery efficiency.

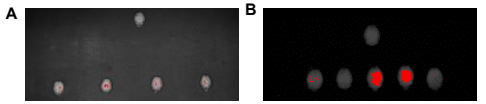


Figure 4. IVIS images of brain distribution of NPLs 24 h post-injection.(**A**) Membrane-coated NPLs.(**B**) Uncoated NPLs.

Conclusions

🧪 **Well-Defined NPL:** SizeAstrocyte membrane-coated NPLs exhibited a size range of 50–100 nm, as confirmed by DLS and TEM analysis.

📈 **Enhanced Cellular:** UptakeCoated NPLs showed significantly higher uptake than uncoated NPLs in glioblastoma, astrocyte, and brain endothelial cell lines.

🛡️ **Favorable Safety:** ProfileNo cytotoxic effects were observed in any of the tested cell lines.

🩸 **Preserved Pharmacokinetics:** Membrane coating did not significantly alter systemic circulation, as confirmed by pharmacokinetic analysis.

🧠 **Brain Distribution Assessed by IVIS:** IVIS imaging enabled visualization of the distribution of membrane-coated and uncoated NPLs.