

# Cationized Extracellular Vesicles for Gene Delivery

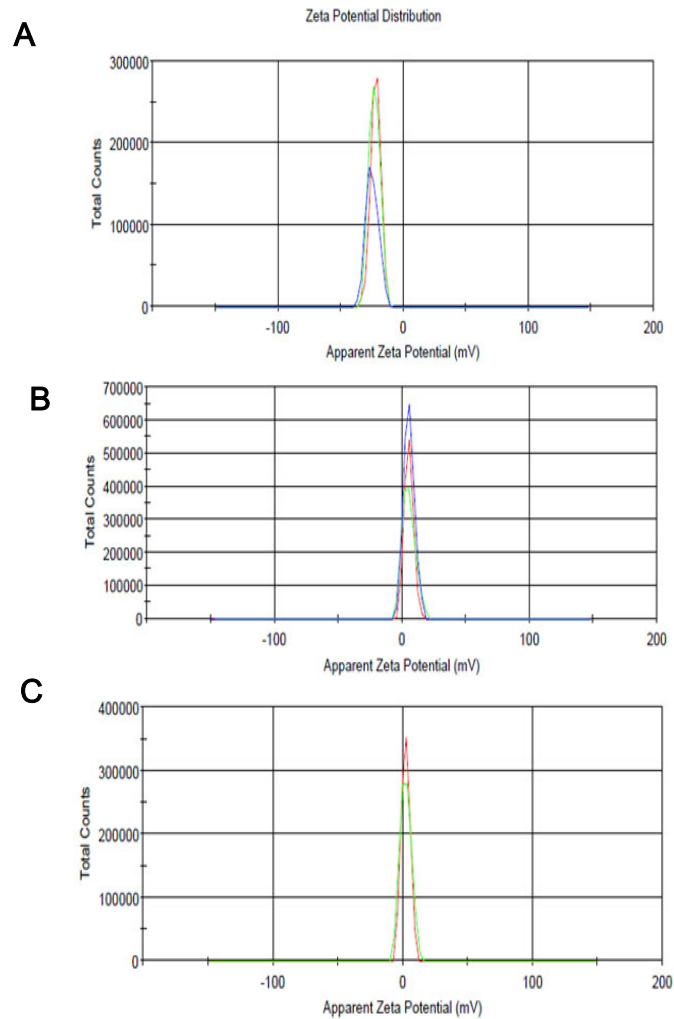
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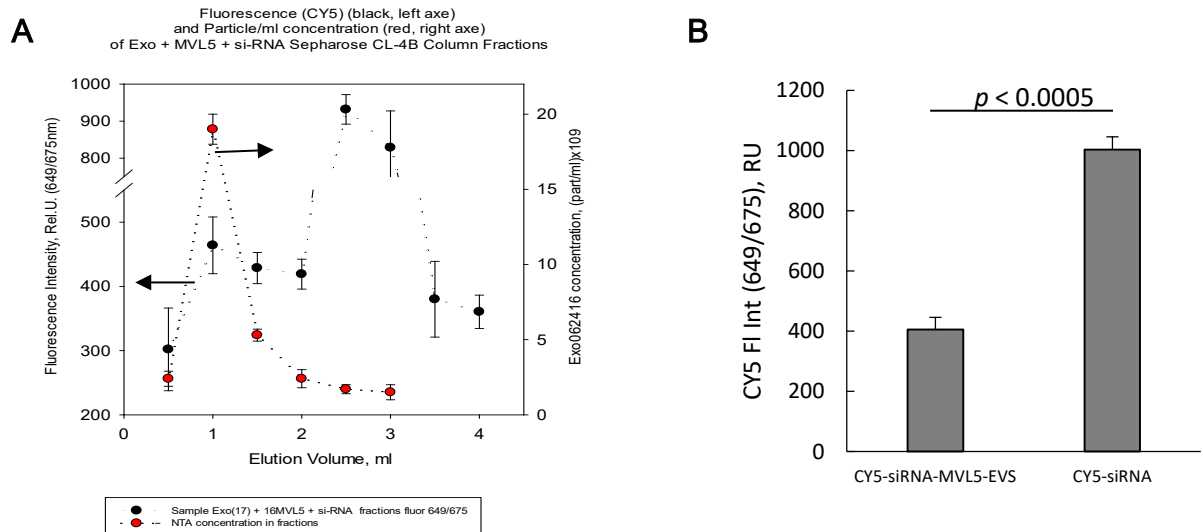
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**Supplementary Data**



**Figure S1. Characterization of modified EVs by DLS.** Zeta potential distribution of naïve EVs (A), MVL5-modified EVs before (B), and after (C) purification from the excess of lipid by PEG precipitation. MVL5-EVs the cationic lipid was added to an aqueous dispersion of macrophage-derived EVs at 25  $\mu\text{g}$  MVL5 *per*  $10^{10}$  of EVs.



**Figure S2. Purification of MVL5-EVs-CY5-siRNA formulations by column chromatography (A) and PEG precipitation (B).** MVL5-EVs was purified from non-incorporated lipid, incubated (1h at 37 °C in siRNA buffer) with fluorescently labeled CY5-siRNA, and then the obtained formulation was purified second time by size-exclusion chromatography (A) or PEG precipitation (B). A portion of siRNA loaded onto EVs was eluted in a void volume (A) or precipitated after PEG addition (B). In both cases, about 40% of all siRNA was loaded into MVL5-EVs. To manufacture MVL5-EVs the cationic lipid was added to an aqueous dispersion of macrophage-derived EVs at 10 µg MVL5 *per* 1x10<sup>10</sup> particles, and then purified from non-incorporated lipid by PEG precipitation. CY5-siRNA (1.2 µM), Sepharose CL-4B column,  $\lambda_{ex}$  = 649 nm;  $\lambda_{em}$  = 675 nm.