Supplementary Materials: Glycan-Modified Melanoma-Derived Apoptotic Extracellular Vesicles as Antigen Source for Anti-Tumor Vaccination

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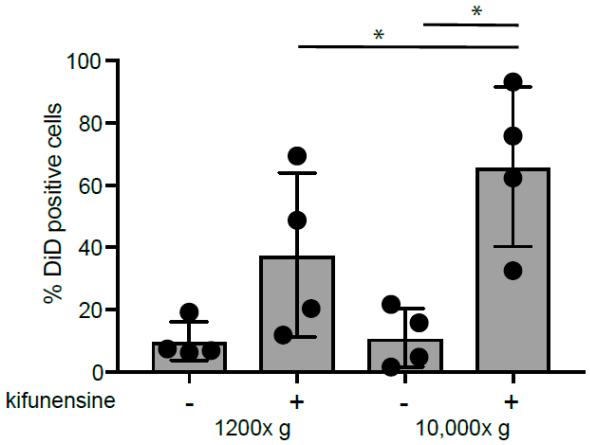

Figure S1. MoDCs favor ApoEVs–HM isolated at 10,000× *g*. MoDCs were incubated with DiD labeled ApoEVs or ApoEVs-HM (50 µg/mL) isolated at 1200× *g* and 10,000× *g* for 45 min at 4 °C, before cells were transferred to 37 °C for 60 min. Uptake was measured by flow cytometry. Data shown as mean ± SD of $n = 4$. Data analyzed with one-way repeated measures ANOVA with Tukey post-hoc test, * $p < 0.05$.

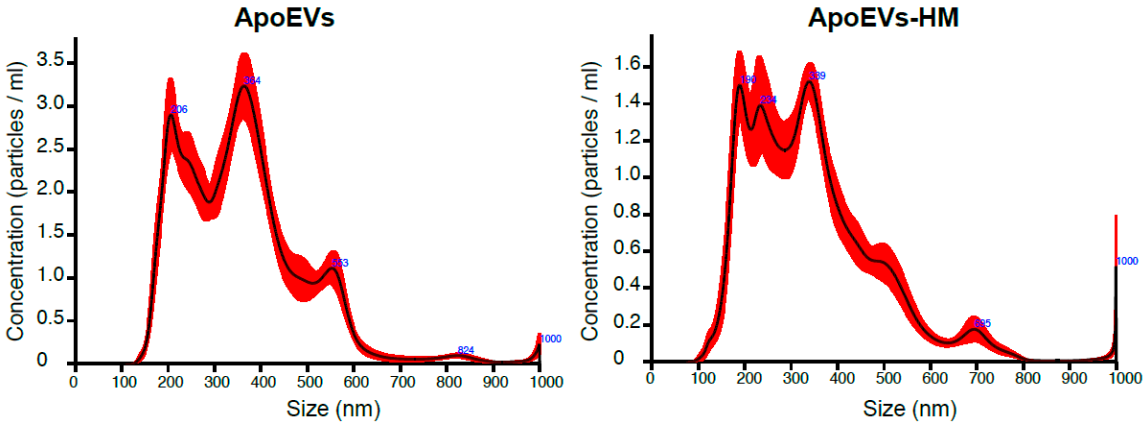

Figure S2. Profile of size distribution of ApoEVs and ApoEVs-HM, measured by nanoparticle tracking analysis (NTA).

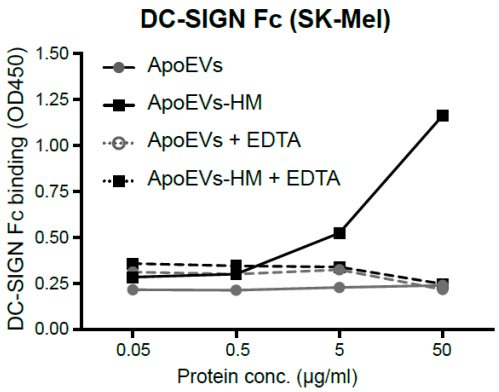

Figure S3. SK-Mel28 derived ApoEVs-HM express DC-SIGN binding ligands. Different concentrations of SK-Mel28-derived ApoEVs were coated on an ELISA plate and DC-SIGN binding was detected by DC-SIGN-Fc staining.

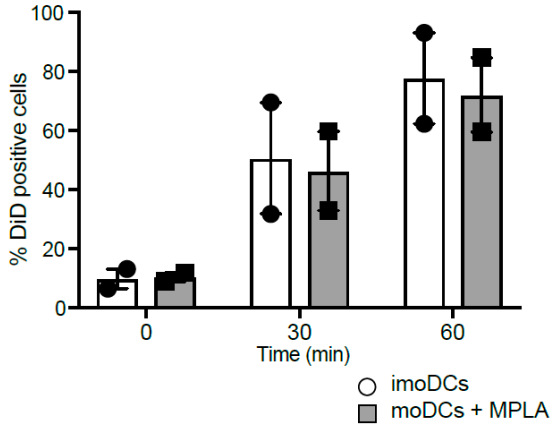

Figure S4. Simultaneous administration of a TLR4-stimulus did not affect ApoEV-HM uptake. Internalization of ApoEVs after 0, 30 and 60 min by TLR4-stimulated moDCs (10 µg/mL MPLA). Data of n=2.

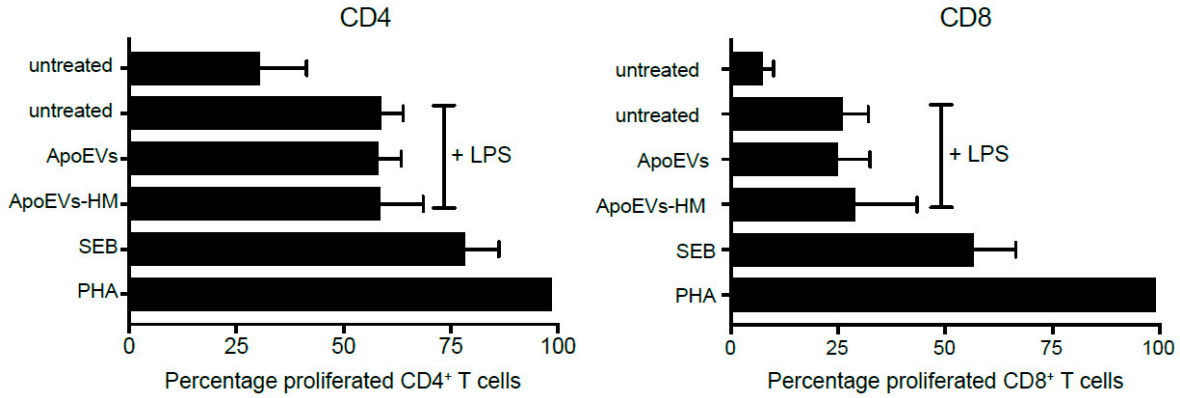

Figure S5. Allogeneic mixed leukocyte reaction (MLR), ApoEV- and ApoEV-HM-loaded (200 µg/mL) and LPS-stimulated moDCs and violet tracer stained peripheral blood lymphocytes (PBLs) were co-cultured for 7 days. T cell proliferation was analyzed by flow cytometry using the violet tracer dilution as a read-out. Data represents the mean + SD and are representative graphs of $n = 2$.

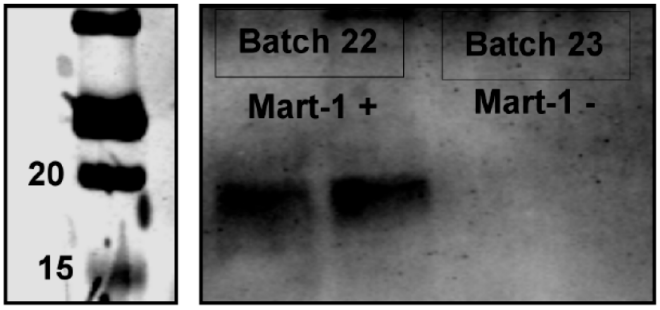

Figure S6. Western blot for the detection of MART-1 protein (18 kDa) in the ApoEVs and ApoEVs-HM. Equal amounts of protein were loaded of lysed ApoEVs and ApoEVs-HM.