pharmaceutics

Supplementary Materials: Mesenchymal Stem Cell Derived Biocompatible Membrane Vesicles Demonstrate Immunomodulatory Activity Inhibiting Activation and Proliferation of Human Mononuclear Cells

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Results

1. Impact of CIMVs uptake efficiency on proliferation of lymphocytes

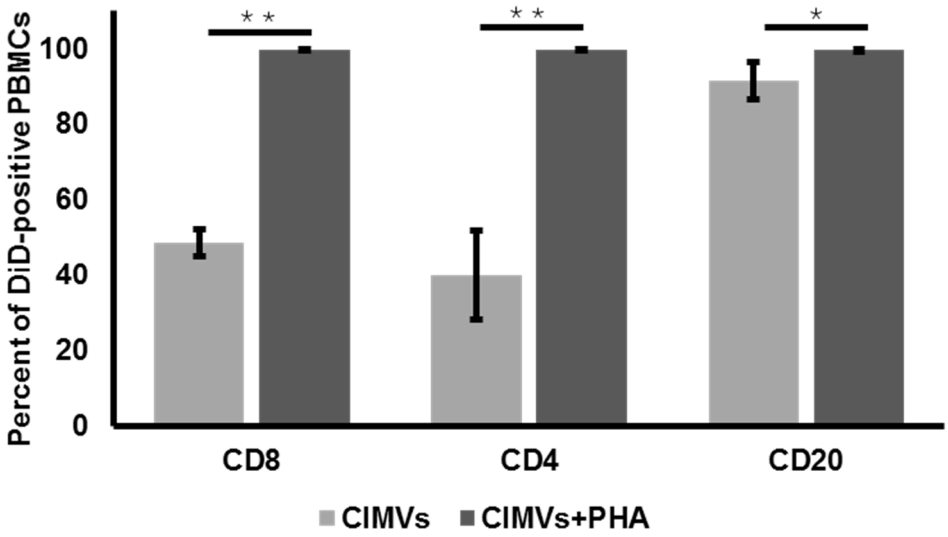

Figure S1. CIMVs-MSCs uptake by lymphocytes. CIMVs-MSCs (10 μg) stained with membrane dye DiD were incubated with PBMCs for 24 h, followed by treatment with PHA (10 μg/mL). PBMCs were analyzed using flow cytometer BD FACS Aria III (BD Bioscience, USA). The data represent mean ± SD. *p-value ≤ 0.05, **p-value ≤ 0.01.

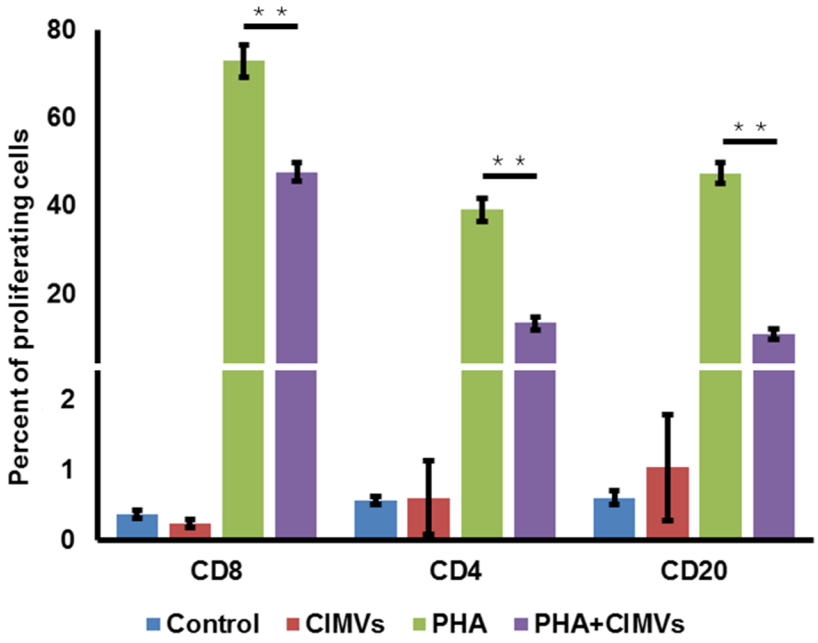

Figure S2. CIMVs-MSCs effect on PHA-induced proliferation of T-cytotoxic (CD8+), T-helper (CD4+), and B-cells (CD20+). Lymphocytes were stained with CFDA SE, followed by incubation with DiD-labeled CIMVs-MSCs for 24 h and treatment with PHA (10 μg/mL). The percent of proliferating cells was evaluated 3 days after PHA incubation. Data represent mean ± SD. ***p*-value ≤ 0.01.

2. FACS analysis of murine PBMCs: a gating strategy

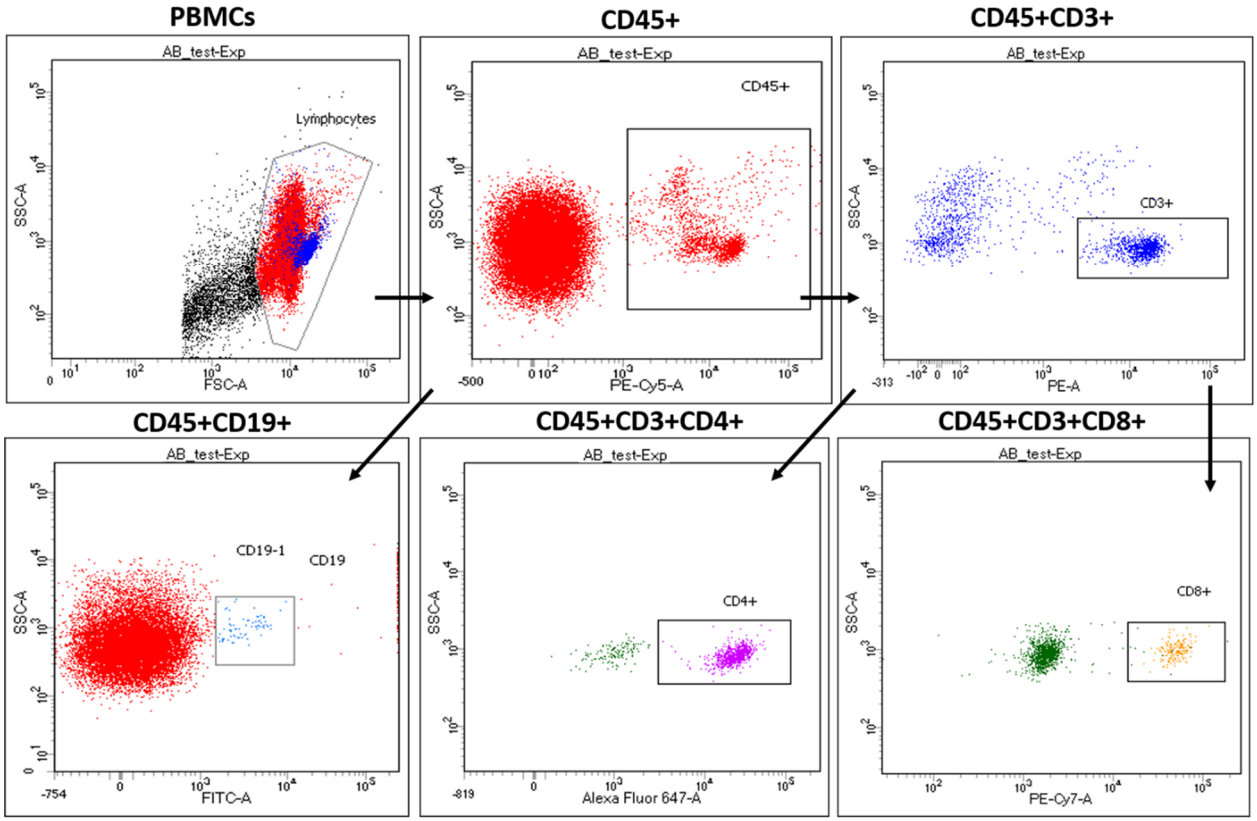

Figure S3. Analysis of murine PBMCs using flow cytometry. PBMCs were incubated with anti-CD45, anti-CD3, anti-CD4, anti-CD19, and anti-CD8 monoclonal antibodies and analyzed using flow cytometer BD FACS Aria III (BD Bioscience, USA). Histograms were built using BD FACSDiva 8 software (BD Bioscience, USA).

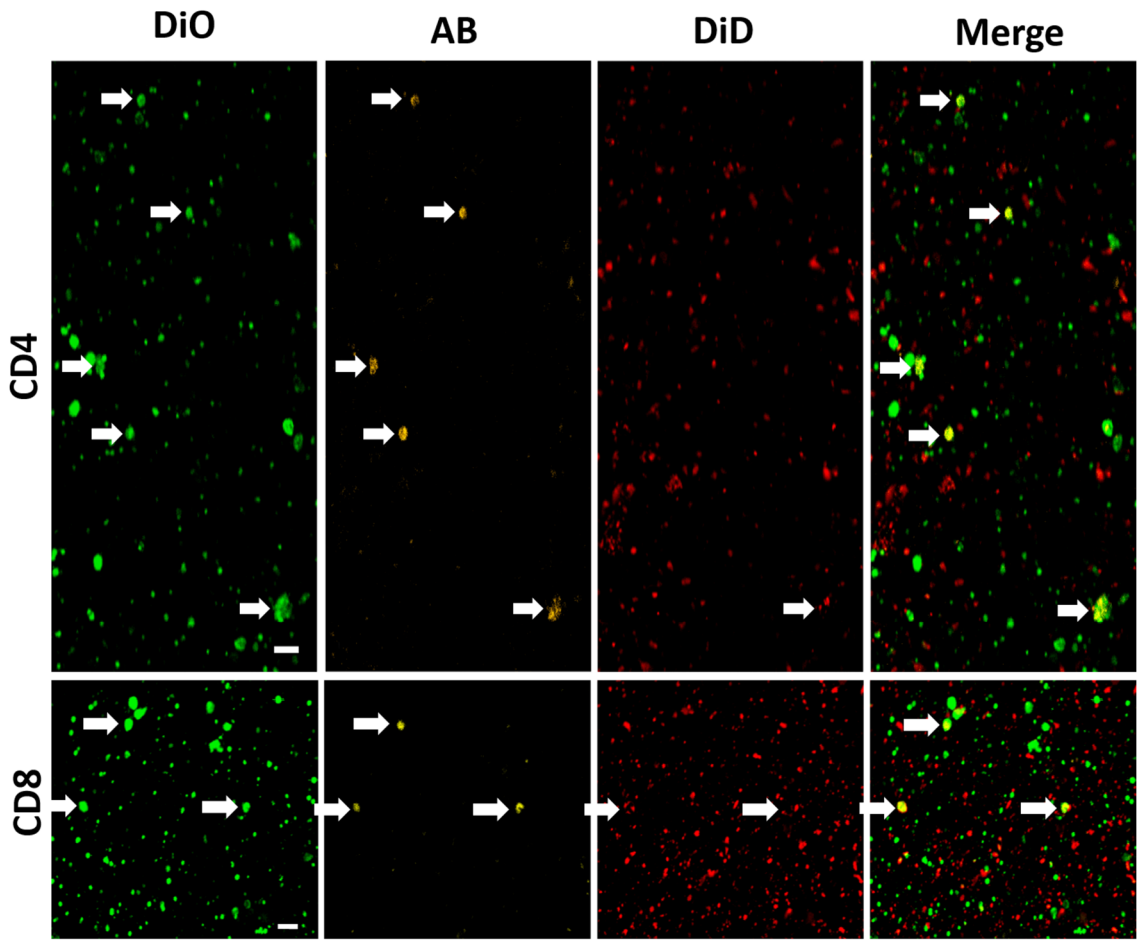

Figure 4. Analysis of co-expression of DiD-stained CIMVs with CD4-positive and CD8-positive cell populations. ⇨ co-localization of DiD and AB-staining signals. Scale bar 20 μm.

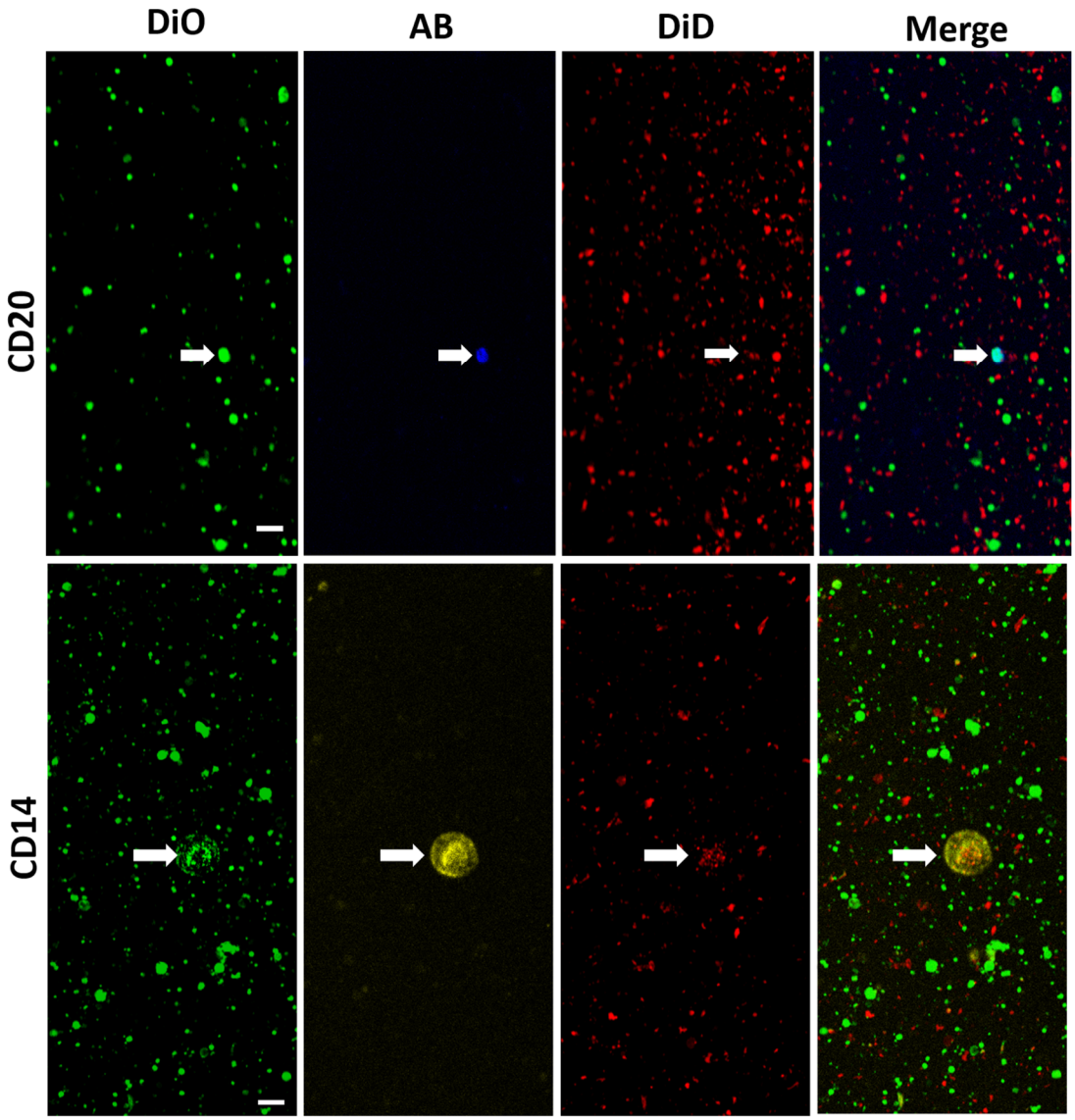

Figure S5. Analysis of co-expression of DiD-stained CIMVs with CD20-positive and CD14-positive cells populations. ⇨ co-localization of DID and AB-staining signals. Scale bar 20 µm.