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Article

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Hardwiring tissue-specific AAV transduction in mice through engineered receptor expression

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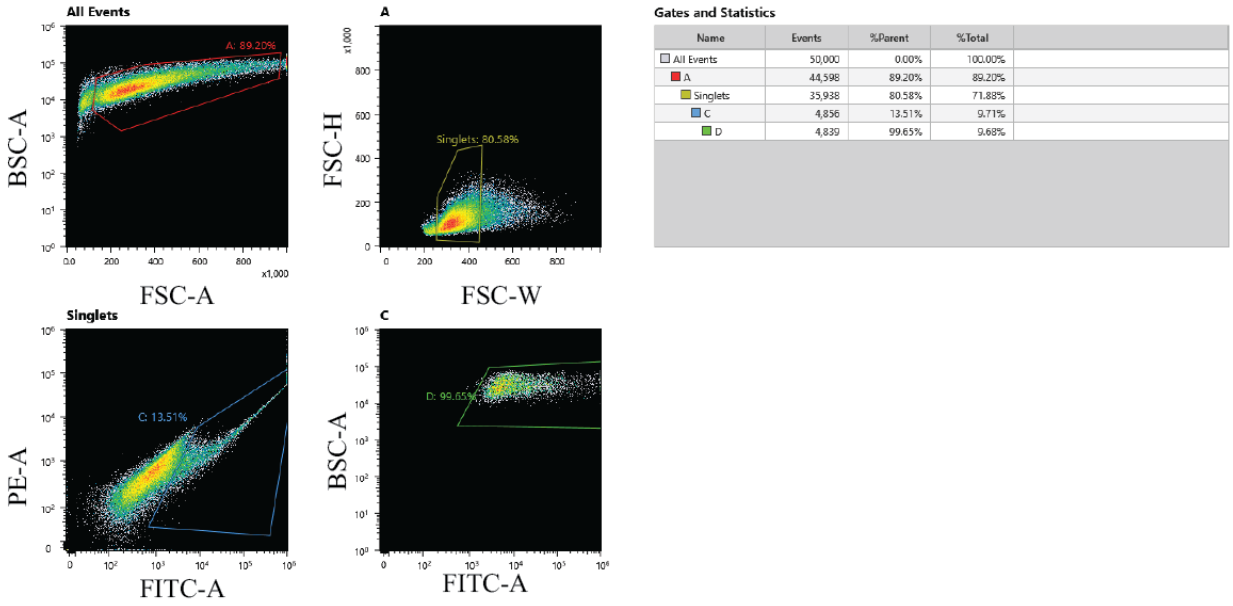
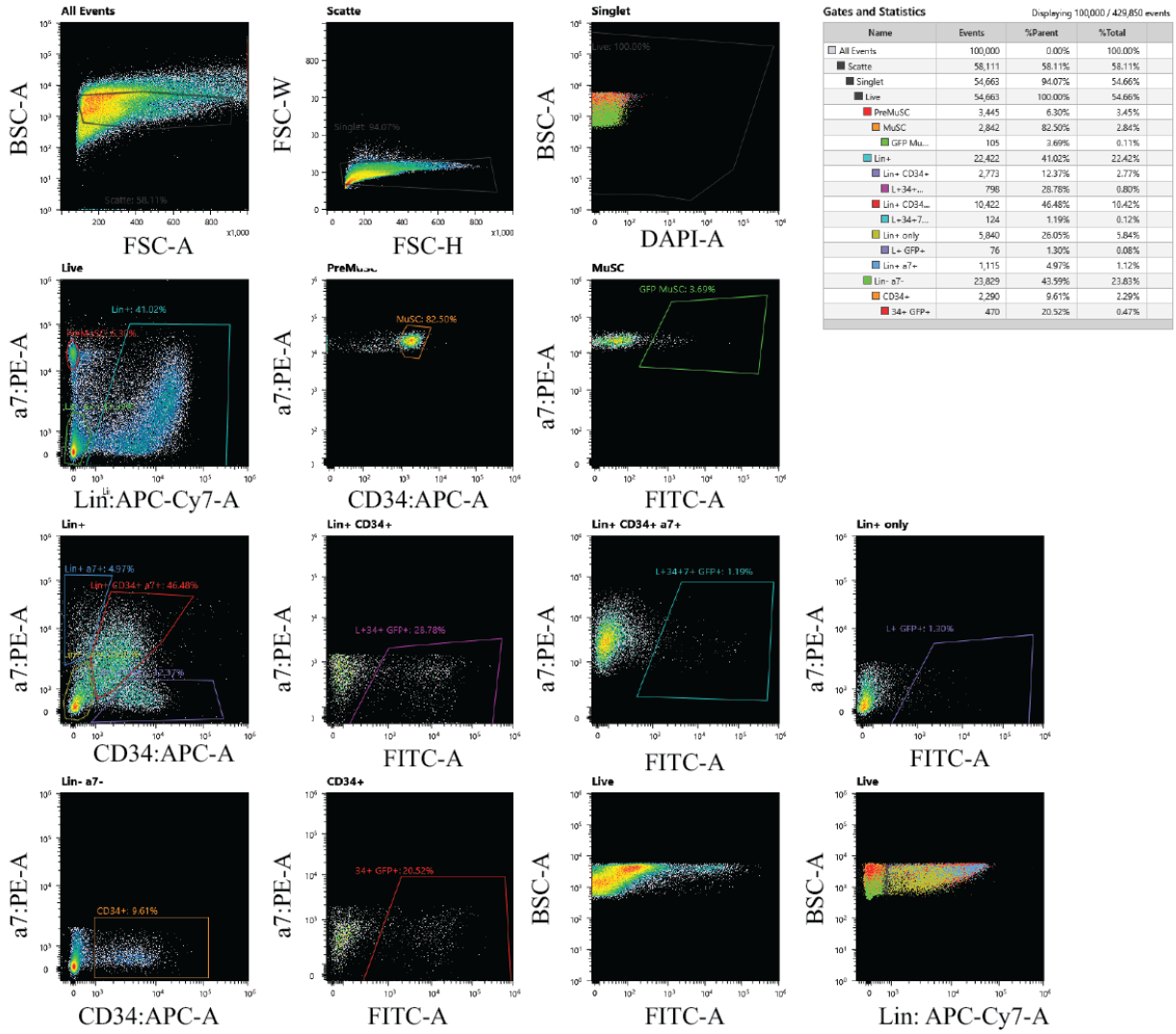
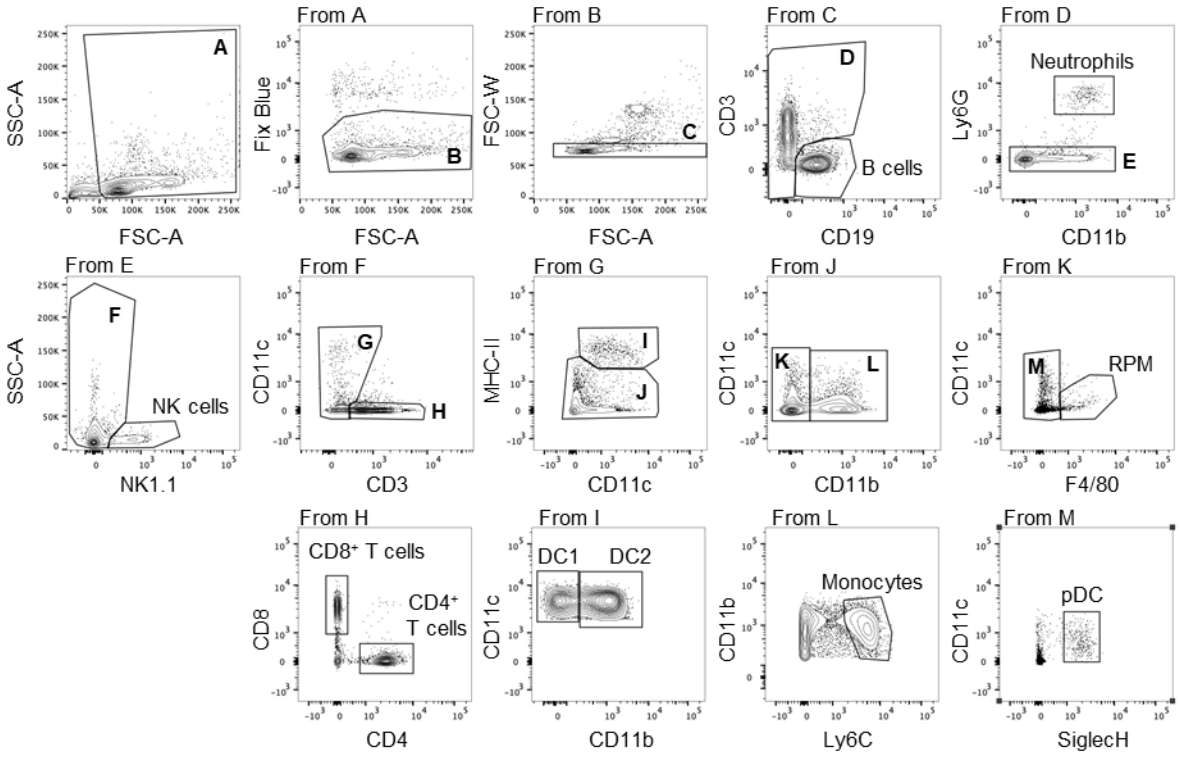


Supplementary Figure 1: Gating strategy for transduction experiments FACS. a, MEFs transduced with AAV9-sgRNA-PCSK9-GFP were sorted to isolate the GFP+ population. FSC and SSC were used to identify single cells and GFP+ cells were gated based on a comparison to untransduced cells.



Supplementary Figure 2: MuSCs were isolated from TA muscle and diaphragm of mice and digested to get a single cell suspension. Live mononuclear cells dissociated from AAV injected muscles were gated by FSC-A vs. SSC-A to remove low and high SSC debris. Then singlets were selected for by gating on FSC-w and FSC-h. Live cells were selected based on low DAPI signal. MuSCs were enriched by gating for a7-PE positivity and lineage marker (CD45-PE-Cy7, CD11b-PE-Cy7, CD31-PE-Cy7) negativity and further selected by gating on CD34-APC positive and SSC-A low populations. Based on previous data, this strategy yields MuSCs at >95% purity based on Pax7+ staining. Antibody gates and compensation were established using unstained, single stained, and FMO controls. GFP gates were established on untransduced cells or tissues.



Supplementary Figure 3: Gating strategy for immune cell identification using FACS. Live cells were gated from the Fix Blue negative population, followed by singlet gating. Cell types were identified as outlined: B-cell (CD19+, CD3-), Neutrophil (CD11b+, Ly6G+), NK cells (NK1.1+), CD8+ T cells (CD3+, CD8+), CD4+ T cells (CD3+, CD4+), DC1 (MHC-II+, CD11c+, CD11b-), DC2 (MHC-II+, CD11c+, CD11b+), Monocytes (CD11b+, Ly6C+), RPM (CD11b-, F4/80+), pDC (F4/80-, Siglec+). Population frequency was identified and reported in Extended Data Fig. 2c. DC populations were combined to simplify reporting of immune cell phenotype.