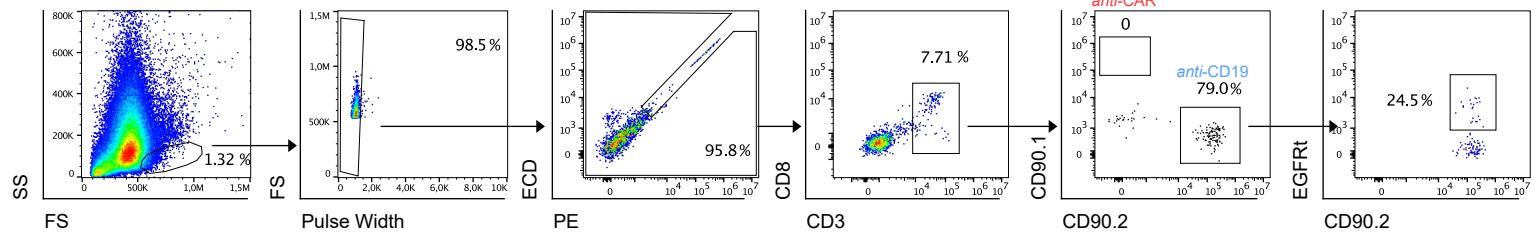
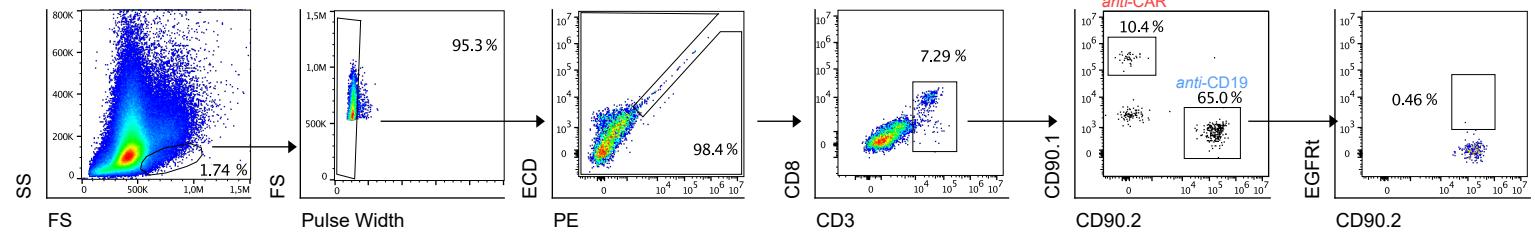


Supplementary Figure 1. Tracking of anti-CAR CAR-T cells *in vivo*. (A) Western blot showing pCD3 ζ expression in rested or activated mock and anti-CAR CAR-T cells. GAPDH was used as a loading control. (B) Schematic of the experimental setup. (C) Representative flow cytometry plots (day 14 post infusion) and quantification over time of mock (CD45.1^{+/−}/EGFRt[−]) and anti-CAR CAR-T cells (CD45.1^{+/−}/EGFRt⁺). (D) Representative flow cytometry plots and quantification of endogenous B cells over time. (E) Representative flow cytometry plots of mock cells and anti-CAR CAR-T cells expressing the activation marker CD69 and the activation/exhaustion markers PD-1 and TIM3. (F) Quantification of PD-1⁺, PD1⁺TIM3⁺ and CD69⁺ transferred T cells in blood over time. (G) Quantification of transferred T cells, endogenous B cells and the activation marker CD69 as well as the activation/exhaustion markers PD-1 and TIM3 in bone marrow, spleen and lymph nodes at the endpoint (day 50). Data are shown as mean + SD. In (C-G), statistical analyses were performed by two-way ANOVA with Sidak's multiple comparisons test for blood, and unpaired t-test for tissues. **p < 0.01, ***p < 0.001, *p < 0.0001.

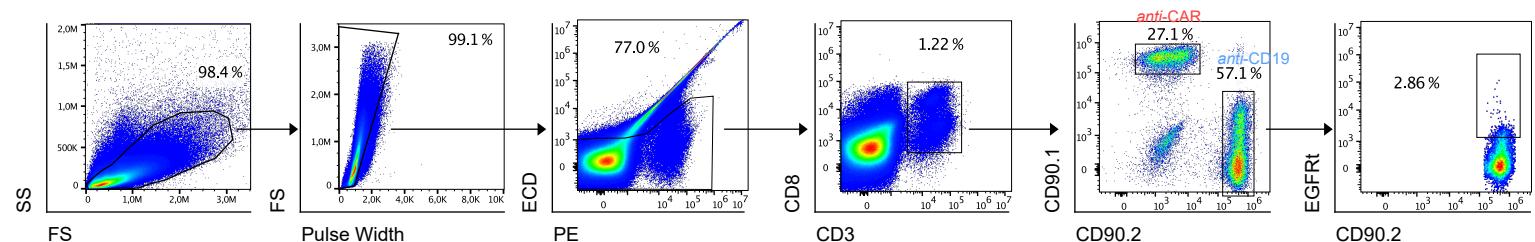
anti-CD19 CAR (blood)



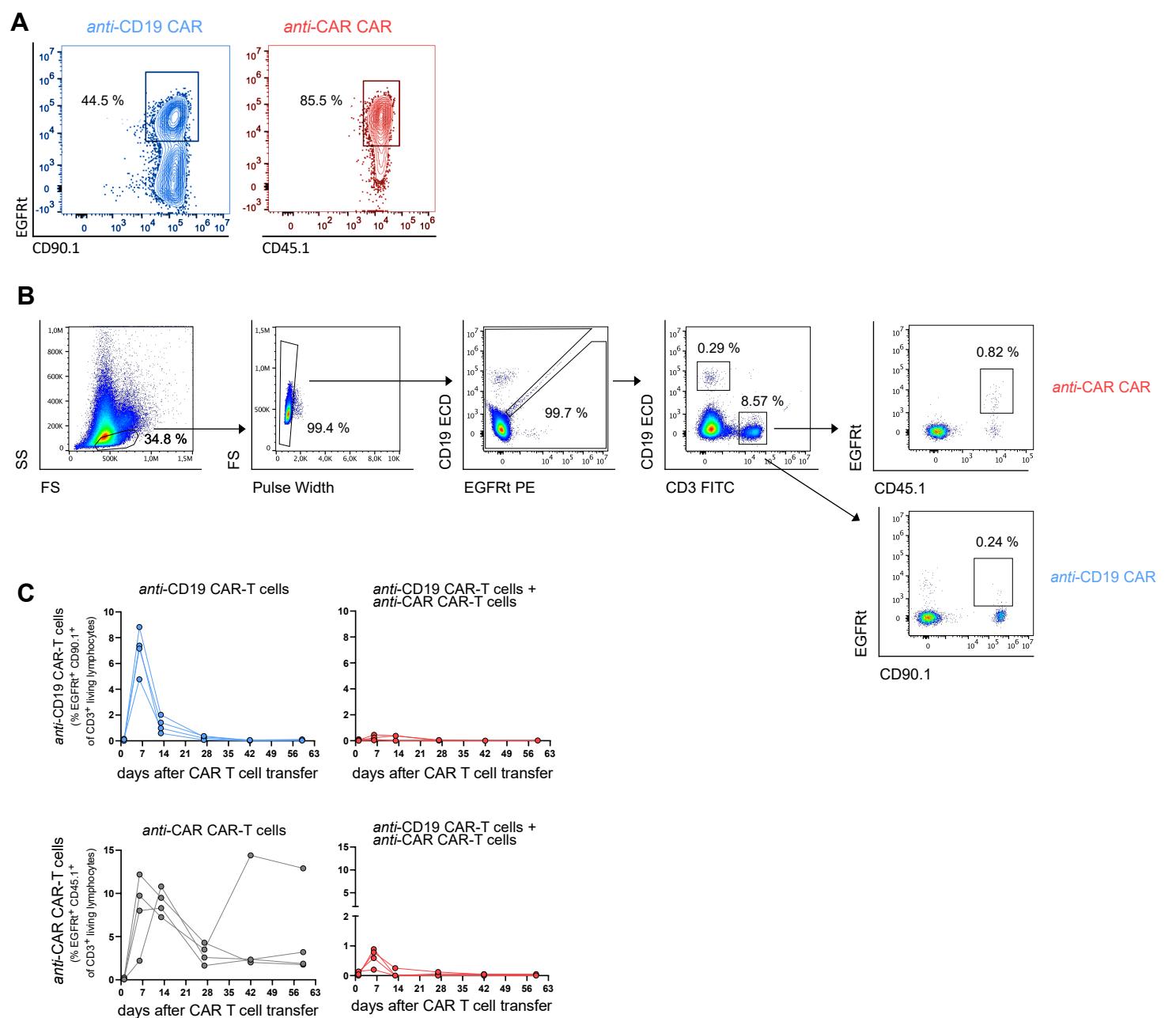
anti-CD19 CAR + anti-CAR CAR (blood)



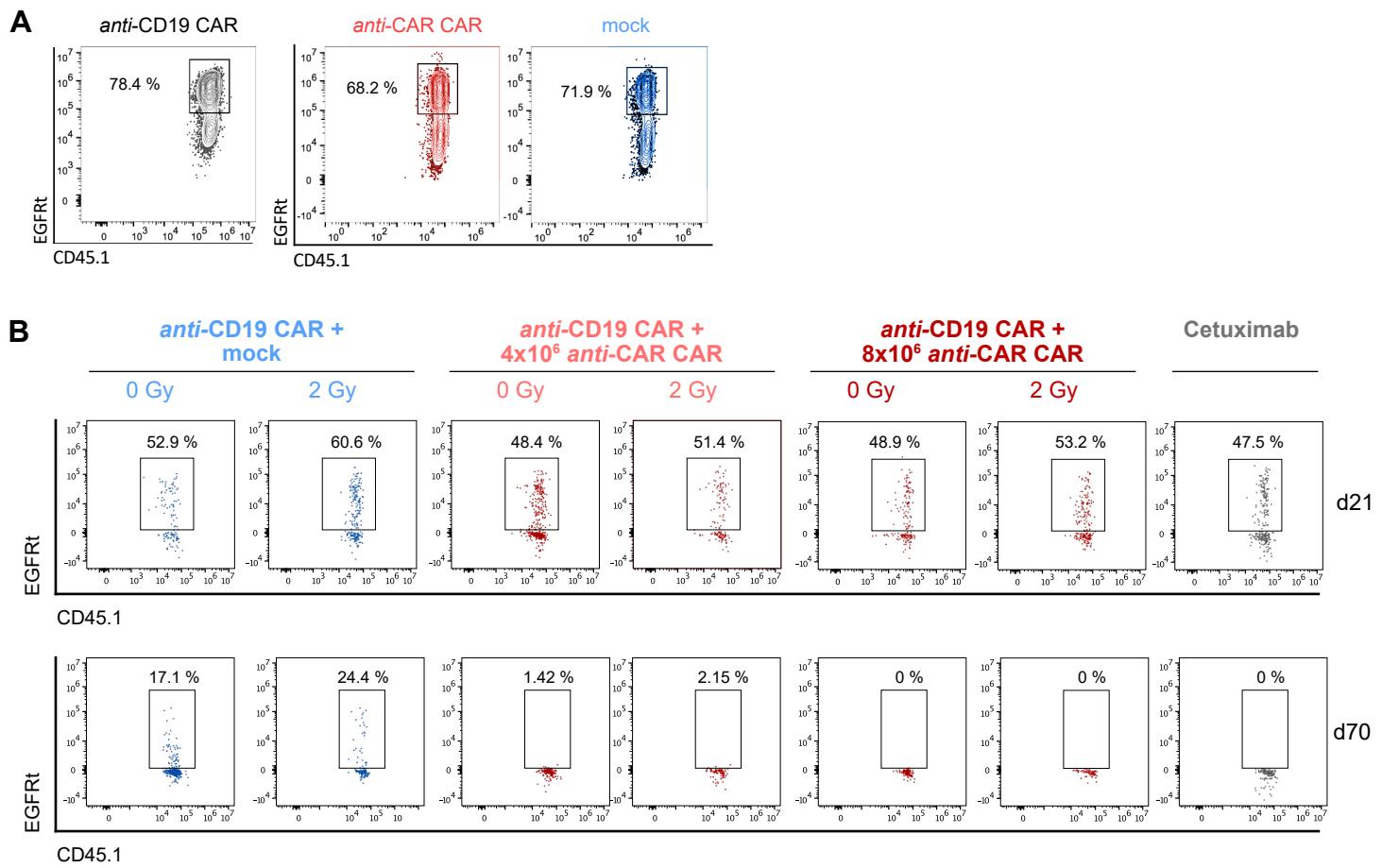
anti-CD19 CAR + anti-CAR CAR (spleen)



Supplementary Figure 2. Tracking of anti-CD19 CAR-T cells. Exemplary flow cytometry plots showing the gating strategy used to assess the persistence of anti-CD19 CAR-T cells ($\text{EGFR}^t \text{ CD90.2}^+$) according to the treatment schedule.



Supplementary Figure 3. Analyses of anti-CD19 and anti-CAR CAR T cells in co-transfer experiments. (A) Cell surface expression of CAR constructs after retroviral transduction. (B) Exemplary flow cytometry plots showing the gating strategy used to evaluate the persistence of anti-CD19 CAR-T cells ($\text{EGFRt}^+ \text{CD90.1}^+$) and anti-CAR CAR-T cells ($\text{EGFRt}^+ \text{CD45.1}^+$). (C) Single mice showing quantification of the frequencies of anti-CD19 CAR-T cells (upper) and anti-CAR CAR-T cells (lower) in blood over time.



Supplementary Figure 4. Analyses of anti-CD19 CAR-T cells in a model of therapeutic administration of anti-CAR CAR-T cells. (A) Cell surface expression of CAR constructs after retroviral transduction. (B) Representative flow cytometry plots showing anti-CD19 CAR-T cells in the peripheral circulation at the indicated time points.