SUPPLEMENTAL MATERIAL

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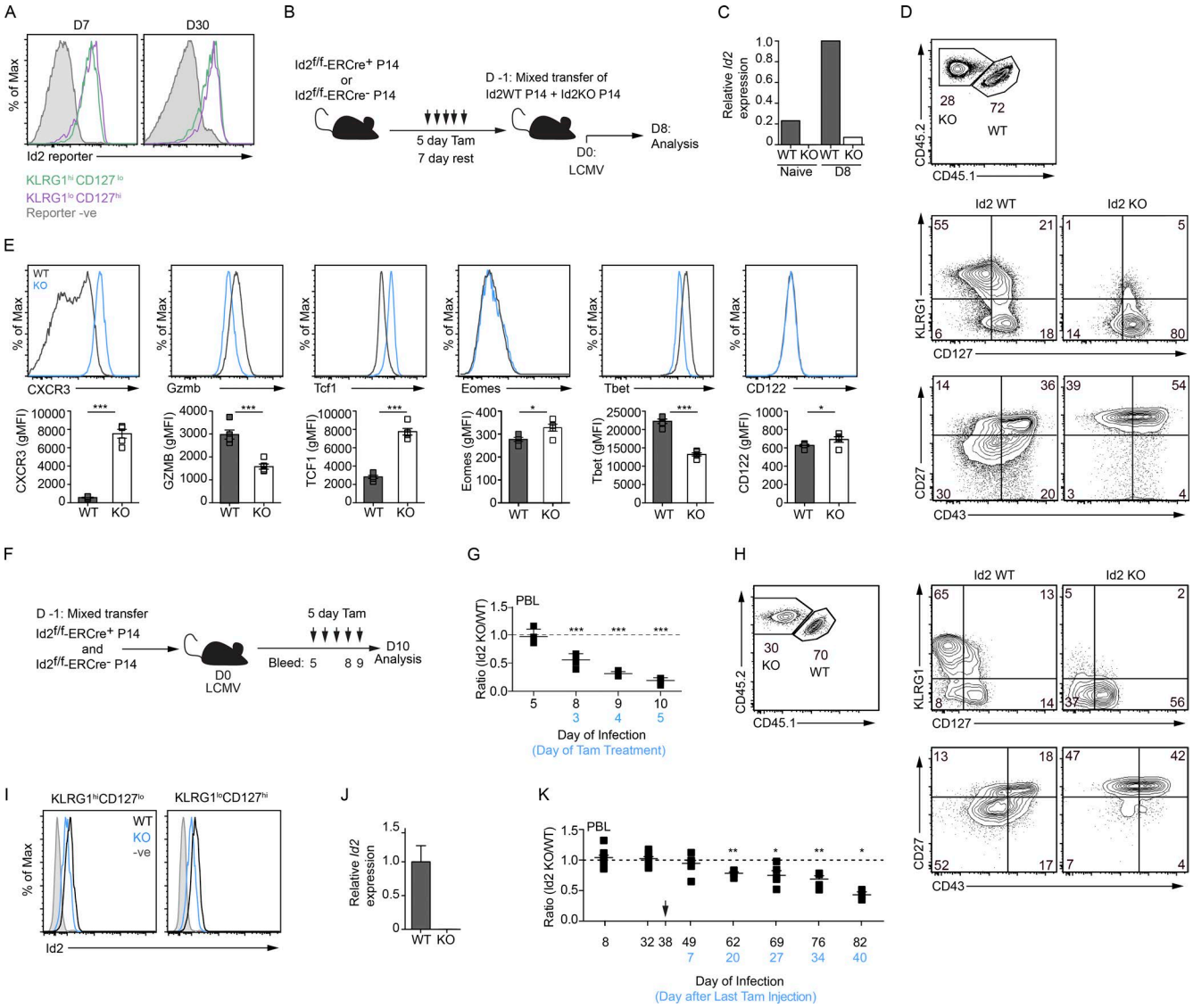

Figure S1. **Id2-deficient cells mount a defective effector CD8⁺ T cell response to LCMV. (A)** Expression of Id2-YFP reporter by KLRG1^{hi}CD127^{lo} or KLRG1^{lo}CD127^{hi} P14 CD8⁺ T cell populations on day 7 or 30 after LCMV infection. **(B)** Schematic of experimental set-up. Id2$^{f/f}$-ERCre^{-} and Id2$^{f/f}$-ERCre^{+} P14 mice were treated for 5 consecutive days with tamoxifen (Tam) to induce *Id2* deletion. Id2$^{f/f}$-ERCre^{-} and Id2$^{f/f}$-ERCre^{+} CD8⁺ T cells are called Id2WT or Id2KO, respectively, after Tam treatment. 8 d after the last Tam treatment, Id2WT and Id2KO CD8⁺ P14 cells were co-transferred into a WT host that was then infected with LCMV. **(C)** Id2WT and Id2KO P14 cells were sorted, and relative *Id2* mRNA expression was assessed by qPCR. **(D)** Flow cytometry of transferred cells from host spleen 8 d after infection. Frequency of Id2WT and Id2KO among P14 CD8⁺ T cells (top), KLRG1 and CD127 expression (middle), and CD27 and CD43 expression (bottom) are shown. Numbers in the plots represent the percentage of cells. **(E)** Expression of indicated proteins in Id2WT or Id2KO donor populations. **(F–H)** Id2$^{f/f}$-ERCre^{-} and Id2$^{f/f}$-ERCre^{+} P14 CD8⁺ T cells were co-transferred into WT hosts that were then subsequently infected with LCMV. Host mice were treated with Tam 5 d after infection to induce *Id2* deletion. (F) Schematic of experimental set-up. (G) The ratio of Id2KO/Id2WT in host PBL was analyzed at the indicated times after infection (black) or after Tam treatment (blue). (H) Flow cytometry of transferred cells from host spleen 10 d after infection. Frequency of Id2WT and Id2KO among P14 CD8⁺ T cells (left), KLRG1 and CD127 expression (top right), and CD27 and CD43 expression (bottom right) are shown. Numbers in plots represent percent cells. **(I–K)** Id2$^{f/f}$-ERCre^{-} and Id2$^{f/f}$-ERCre^{+} P14 CD8⁺ T cells were co-transferred into WT hosts that were then subsequently infected with LCMV. More than 30 d after infection, host mice were treated with Tam to induce *Id2* deletion. (I and J) 15 d after the last Tam treatment, Id2WT and Id2KO P14 CD8⁺ T cells from host spleen were analyzed by flow cytometry for Id2 protein expression (I) or were sorted and relative *Id2* mRNA expression was assessed by qPCR (J). (K) The ratio of Id2KO/Id2WT in host PBL was analyzed at the indicated times after infection (black) or after Tam treatment (blue). Tam treatment began on day 38 of infection. Data are representative of two independent experiments; $n = 3–11$ mice/group. Data are expressed as mean ± SEM. *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$ (two-tailed paired Student's *t* test).

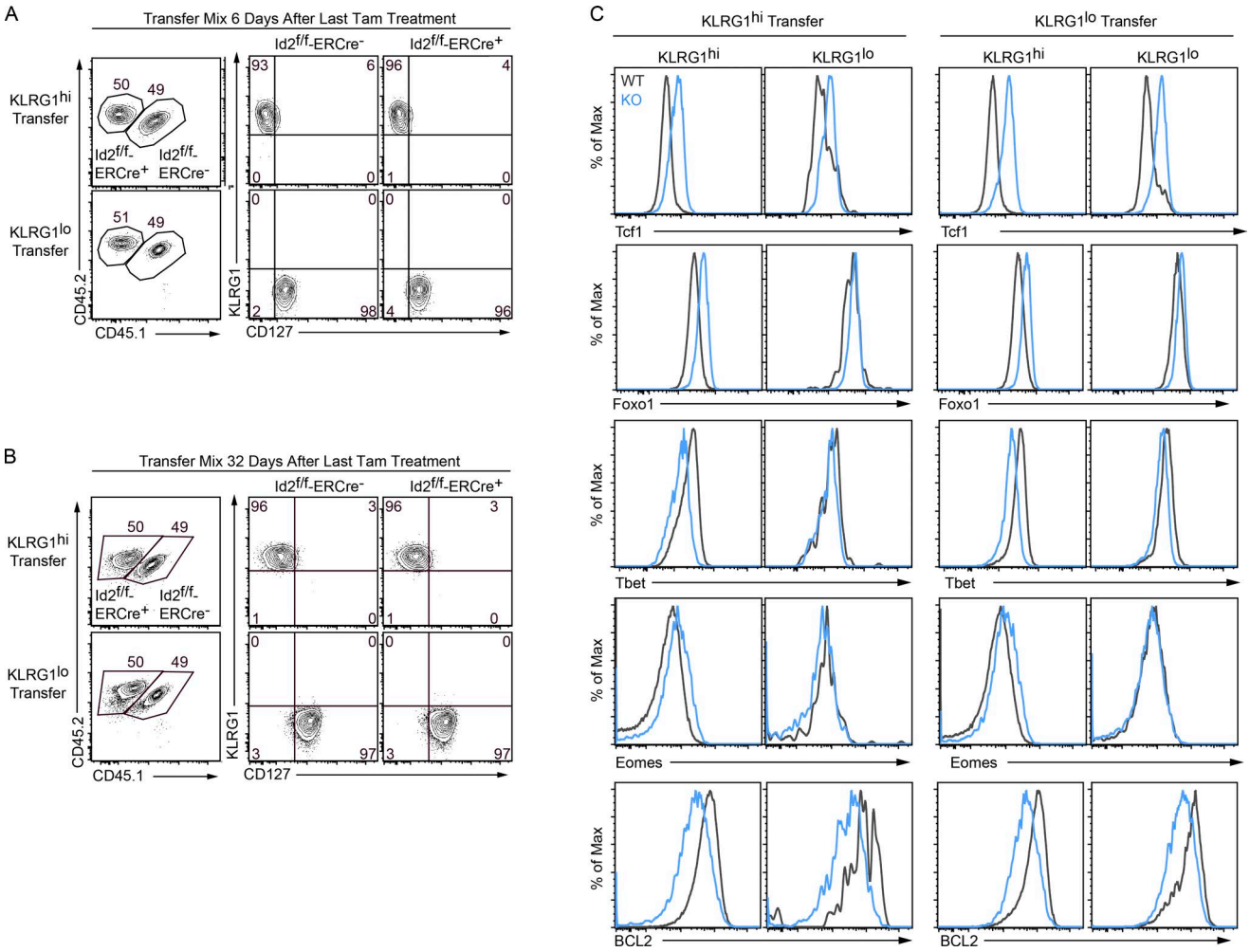

Figure S2. **Phenotype of CD8⁺ T cells in KLRG1^{hi} and KLRG1^{lo} mixes prior to transfer and following rechallenge. (A and B)** Representative flow cytometry analysis for Id2$^{f/f}$-ERCre^{-} and Id2$^{f/f}$-ERCre^{+} KLRG1^{hi} and KLRG1^{lo} transfer mixes from Fig. 3 for CD8⁺ P14 cells assessed 6 d (A) or 32 d (B) after the last tamoxifen (Tam) treatment. **(C)** Hosts that received a mixed transfer of Id2$^{f/f}$-ERCre^{-} and Id2$^{f/f}$-ERCre^{+} KLRG1^{hi} or KLRG1^{lo} P14 CD8⁺ T cells were rechallenged with LCMV on day 6 after the last Tam treatment, as in Fig. 3. Expression of indicated proteins on day 9 of infection on KLRG1^{hi} or KLRG1^{lo} Id2WT or Id2KO donor populations directly ex vivo. Data shown are representative of two independent experiments; *n* = 2–4 mice per group.

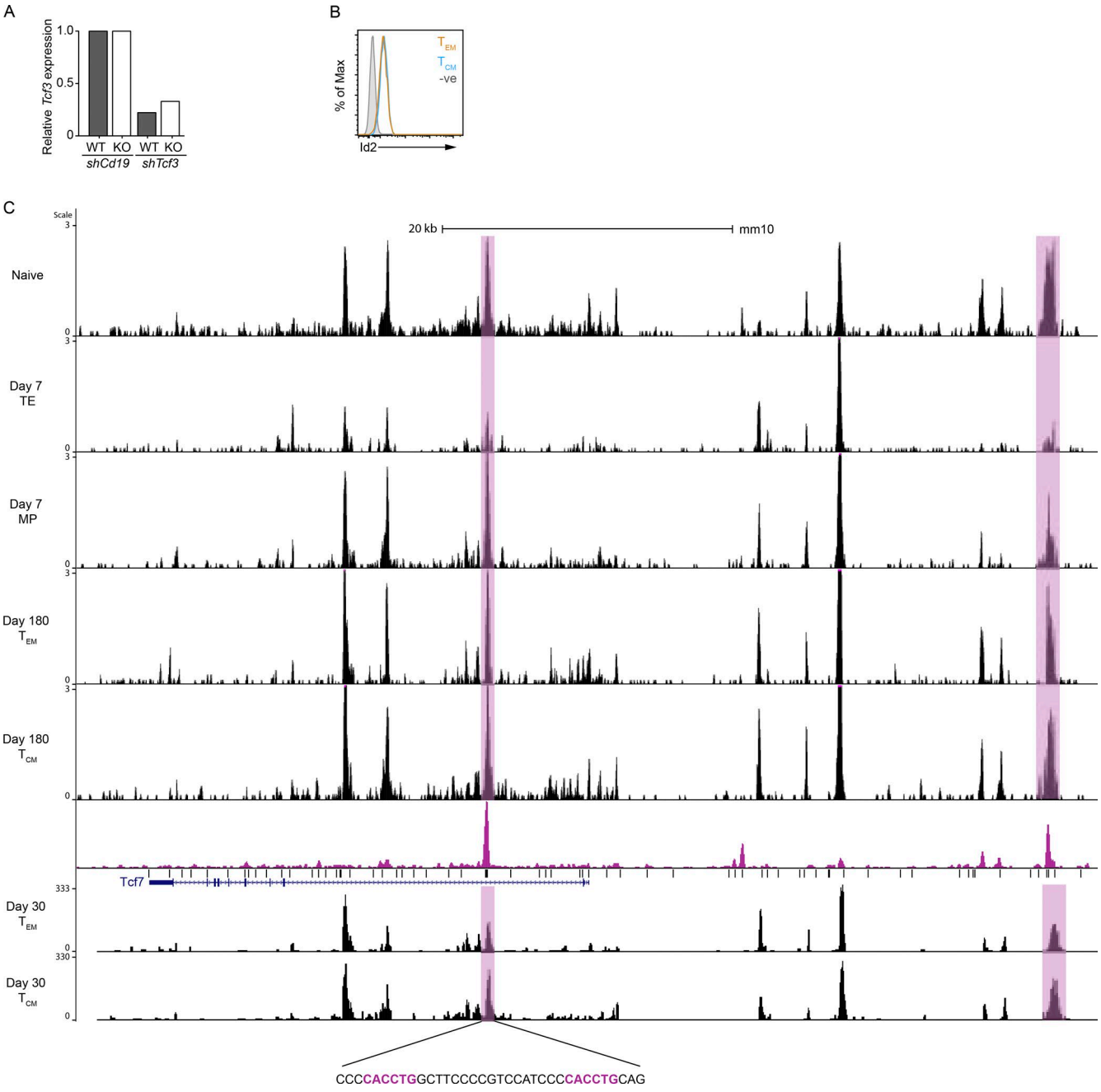

Figure S3. **Zeb2 may regulate E-protein activity in effector-like CD8⁺ T cell populations. (A)** Id2$^{f/f}$-ERCre^{-} and Id2$^{f/f}$-ERCre^{+} P14 CD8^{+} T cells were transduced with the indicated shRNAmir-retroviral supernatant and expanded in vitro for 4 d. Graph indicates relative mRNA expression of *Tcf3* by ametrine^{+} T cells normalized to the sh*Cd19* controls. **(B)** Id2 expression in T_{EM} (CD62L^{-}) and T_{CM} (CD62L^{+}) P14 CD8^{+} T cells at >30 d of infection. **(C)** ATAC-Seq analysis of the *Tcf7*loci for naive, TE (KLRG1^{hi}CD127^{lo}), MP (KLRG1^{lo}CD127^{hi}), T_{EM} (CD62L^{-}), and T_{CM} (CD62L^{+}) P14 CD8^{+} T cells at day 0, 7, 30, or 180 of LCMV infection as indicated. E2A–ChIP-Seq data are aligned to show E-protein binding sites (purple highlight). DNA sequence defines potential Zeb2 binding location with tandem E-box sites in purple. Data are representative of one (A) or two (B) independent experiments; *n* = 4–5 mice per group.