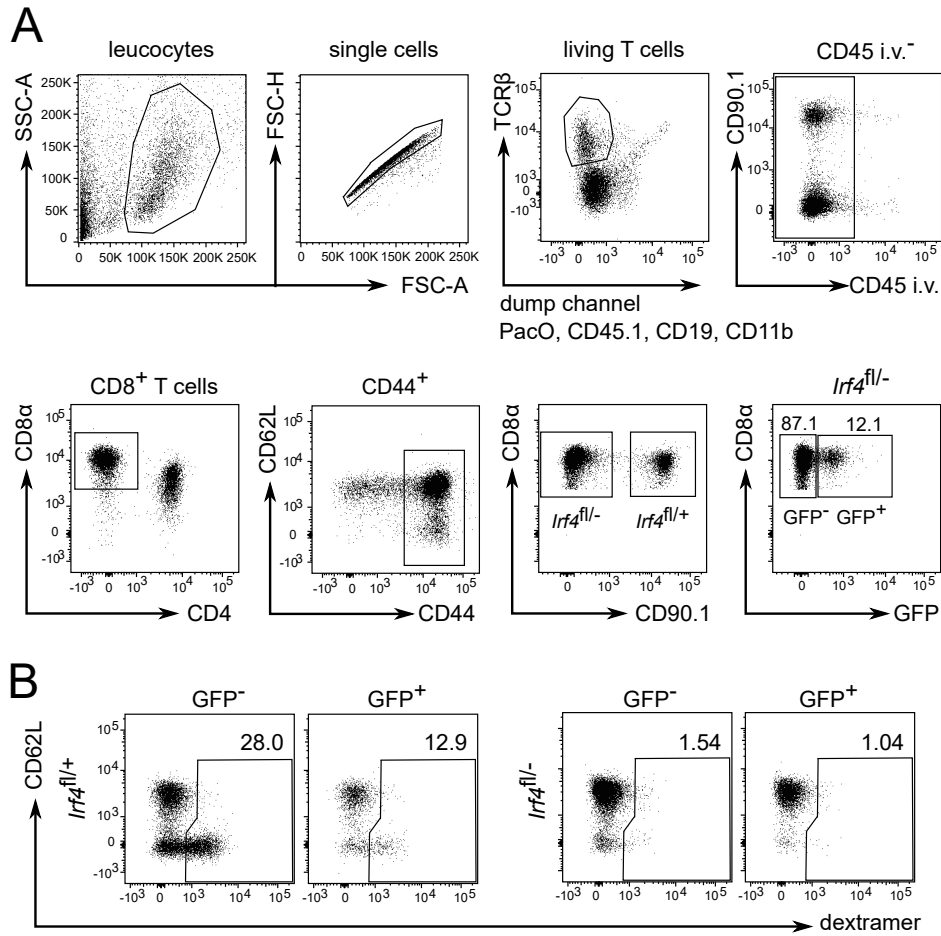


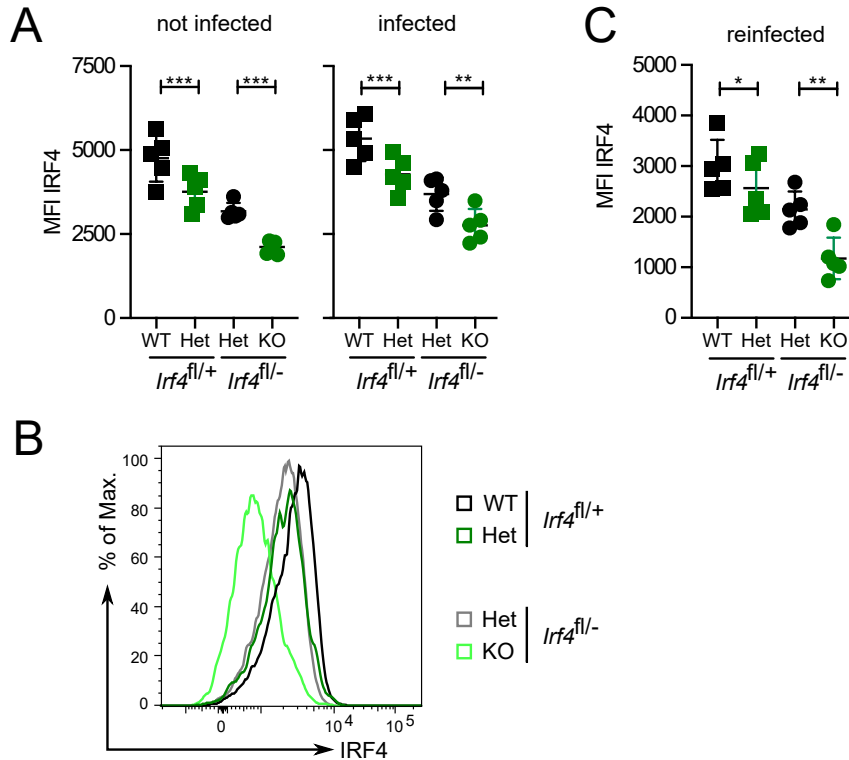
**Suppl. Figure 1. *In vitro* response of IRF4-deficient CD8<sup>+</sup> T cells.**

A) Schematic outline for experiments in Figures 1F and G. B) Spleen cells from *Irf4*<sup>+/+</sup>, *Irf4*<sup>+/-</sup> and *Irf4*<sup>-/-</sup> mice were mixed at 1:1:1 ratio, stimulated with anti-CD3 mAb, anti-CD28 mAb and IL-2. After 3 days, cells were washed and cultured for further 4 days with IL-7. Percentage of *Irf4*<sup>+/+</sup>, *Irf4*<sup>+/-</sup> and *Irf4*<sup>-/-</sup> of CD8<sup>+</sup> T cells. Combined results of 2 experiments with pooled cells from 2 mice per genotype, mean ± SEM. C) Schematic outline for experiments in Figures 1H and I. D, E) CD8<sup>+</sup> T cells from *Irf4*<sup>fl/+</sup> × CreER<sup>T2</sup> and *Irf4*<sup>fl/-</sup> × CreER<sup>T2</sup> mice were treated as described in (C). D, E) CD44 was measured by antibody staining and FACS on days 0, 3 and 23. E) At day 23, IFN-γ and TNF-α were determined by intracellular cytokine staining after stimulation with PMA/ionomycin for 4h. D, E) Representative result of 2 independent experiments with pooled cells from 4 mice per genotype. F, G) CD8<sup>+</sup> T cells were purified from *Irf4*<sup>+/+</sup>, *Irf4*<sup>+/-</sup> and *Irf4*<sup>-/-</sup> mice. Cells were activated with anti CD3 mAb, anti-CD28 mAb and IL-2 for 3 days, and then incubated with IL-7 for 3 weeks. F) On indicated days, CD44 expression was determined. G) IFN-γ and TNF-α were determined after stimulation with PMA/ionomycin for 4h. F, G) Representative result of 3 independent experiments with pooled cells from 4 mice per genotype.



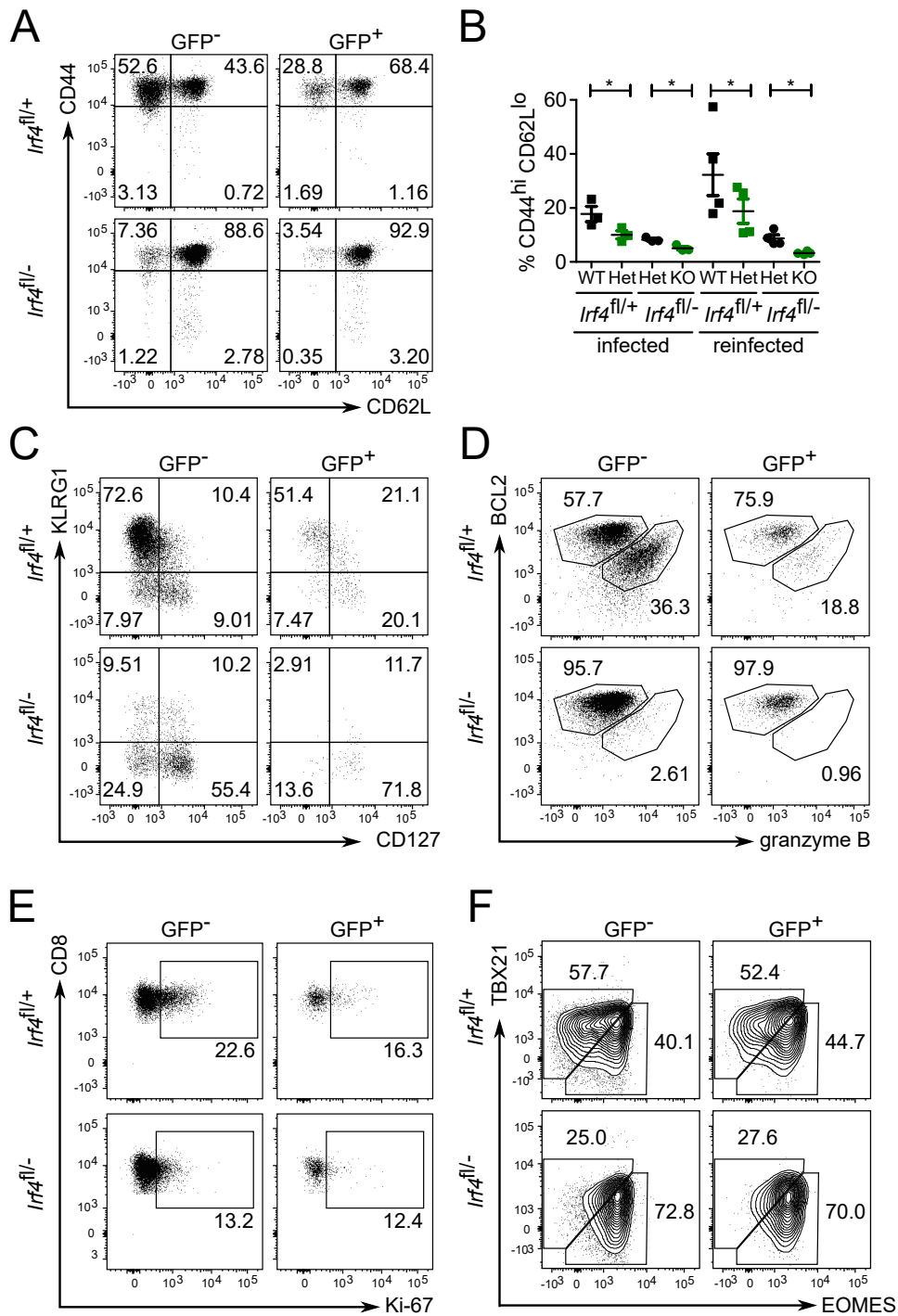
**Suppl. Figure 2. Gating strategies.**

A) Gating strategy for the identification and characterization of CD45.2<sup>+</sup> CD8<sup>+</sup> T cells from *Irf4*<sup>fl/+</sup> × CreER<sup>T2</sup> and *Irf4*<sup>fl/-</sup> × CreER<sup>T2</sup> donors in CD45.1<sup>+</sup> *Rag1*<sup>-/-</sup> recipients. Single leucocytes were defined by their FSC/SSC profile and TCRβ<sup>+</sup> T cells were selected. In some experiments, mice were i.v. injected with fluorochrome-labelled anti-CD45 mAb 3 minutes before killing. Parenchymatic cells were identified by the absence of anti-CD45 staining (CD45i.v.<sup>-</sup>). CD90.1<sup>+</sup> *Irf4*<sup>fl/+</sup> × CreER<sup>T2</sup> CD8<sup>+</sup> T cells were distinguished from CD90.1<sup>-</sup> *Irf4*<sup>fl/-</sup> × CreER<sup>T2</sup> CD8<sup>+</sup> T cells. B) Representative dot plots for dextramer staining of CD8-gated cells in Figure 5A.

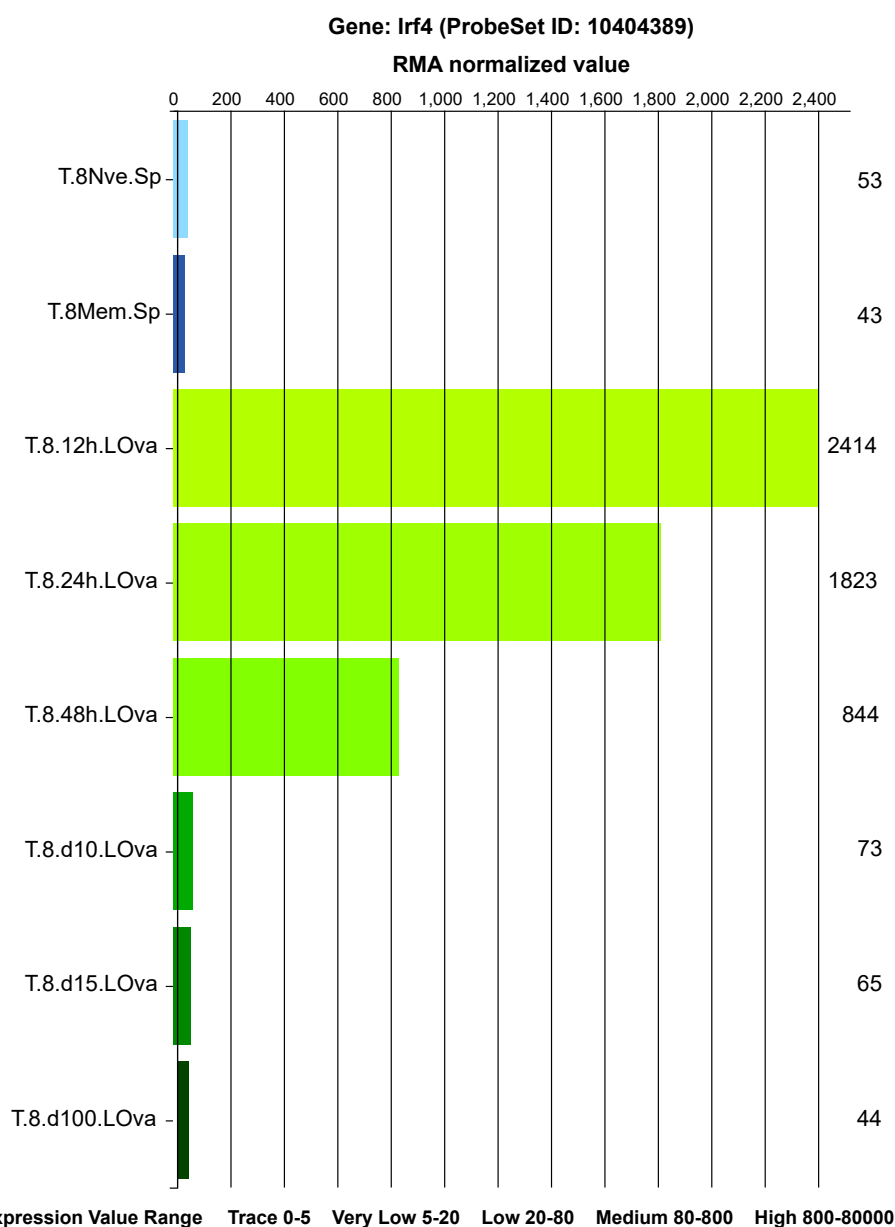


**Suppl. Figure 3. IRF4 Expression *in vivo* after tamoxifen treatment.**

A)  $Rag1^{-/-}$  mice were reconstituted with T cells from naïve  $Irf4^{fl/+} \times CreER^{T2}$  and  $Irf4^{fl/-} \times CreER^{T2}$  mice. Recipient mice were infected with LmOVA. Five weeks later, mice were treated with tamoxifen for 5 days. 24 days after tamoxifen treatment,  $CD8^{+}$  T cells were analyzed (see Figure 2C). Mean fluorescence intensity (MFI) of IRF4 was determined by FACS after stimulation with PMA/ionomycin for 4h. B, C)  $Rag1^{-/-}$  mice were reconstituted with T cells from naïve  $Irf4^{fl/+} \times CreER^{T2}$  and  $Irf4^{fl/-} \times CreER^{T2}$  mice. Recipient mice were infected with LmOVA. Five weeks later, mice were treated with tamoxifen for 5 days. Two weeks after tamoxifen treatment, mice were reinfected with LmOVA. Five days later,  $CD8^{+}$  T cells were analyzed (see Figure 4A). Cells were stimulated for 4h with PMA/ionomycin and IRF4 expression was analyzed by FACS. B) representative results for IRF4 staining. C) MFI of IRF4. Representative results of 3 experiments with 5 - 8 mice in infected groups and 4 - 5 mice in not infected groups (A), and of 3 experiments with 4 - 6 mice in (B) and (C). Mean  $\pm$  SEM, paired t test.



**Suppl. Figure 4. Frequencies of CD8<sup>+</sup> effector T cells and representative staining profiles.** *Rag1*<sup>-/-</sup> mice were reconstituted and treated as described in Figure 4A. Representative dot plots for CD62L and CD44 (A), for KLRG1 and CD127 (C), for granzyme B and BCL2 (D), for Ki-67 (E) and for EOMES and TBX21 (F) of GFP<sup>+</sup> and GFP<sup>-</sup> *Irf4*<sup>fl/+</sup> × CreER<sup>T2</sup> and *Irf4*<sup>fl/-</sup> × CreER<sup>T2</sup> CD8<sup>+</sup> T cells of reinfected mice. B) %-values of CD44<sup>hi</sup>CD62L<sup>lo</sup> CD8<sup>+</sup> T cells of reinfected mice and mice without reinfection (infected). Mean ± SEM, paired t test. A-F) Representative results of 3 independent experiments with 4 - 6 mice in reinfected groups and 2 - 4 mice in infected groups.



**Suppl. Figure 5. Expression of *Irf4* mRNA in different CD8<sup>+</sup> T-cell subsets.**

Expression of *Irf4* in different CD8<sup>+</sup> T-cell populations quantified by micro array analysis. Data are derived from the ImmGen consortium database (Heng TS, et al., Immunological Genome Project Consortium. Nat Immunol. 2008 10: 1091.). From top to bottom: Naïve CD8<sup>+</sup> T cells (CD62L<sup>hi</sup> CD44<sup>lo</sup>), CD8<sup>+</sup> memory T cells (CD44<sup>hi</sup> CD122<sup>hi</sup>), transferred OT1 CD8<sup>+</sup> T cells 12h, 24h, 48h, 10 days, 15 days and 100 days after LmOVA infection. All cell populations were sorted from spleens of naïve C57BL/6J mice or from CD45.1<sup>+</sup> C57BL/6J mice that had received ovalbumin-specific CD8<sup>+</sup> T cells from CD45.2<sup>+</sup> OT1 transgenic mice and were infected with LmOVA.