SUPPLEMENTAL DATA

Supplemental Figure 1 - related to Figure 1. Effect of IFNβ on CD4 Th differentiation, PD-L1 expression on dendritic cells, and LCMV titers after antibody treatment following late priming of virus-specific CD4 T cells.

A. WT and IFNβ-/- mice were CD8 depleted prior to infection with LCMV Cl-13 to prevent any difference in virus titers due to loss of IFNβ antiviral activity. CD4 SMARTA T cells were transferred at day 21 after LCMV-Cl13 infection into WT, IFNβ-/- or WT mice that had been treated with anti-IFNAR blocking antibody prior to cell transfer. Flow plots depict the frequency and bar graphs the numbers of splenic Th1 ($SLAM^{hi}$ $CXCR5^{lo}$) and Tfh ($SLAM^{lo}$ $CXCR5^{hi}$) CD4 SMARTA T cells 8 days after transfer.

B. Virus-specific CD4 NIP transgenic cells were transferred into mice 1 hour prior to LCMV-Cl13 infection (Early priming) or into mice infected 21 days earlier with LCMV-Cl13 infection (Late priming). All mice used for late priming were CD4 depleted prior to LCMV infection to keep virus high and prevent control of infection by blocking antibodies. One day prior to late priming, mice were treated with either isotype control or anti-PD-L1 and IL-10R (dual block - DB) blocking antibodies. Antibody treatment continued on day 1, day 4 and day 7 post priming. Bar graphs depict number of Th1 and Tfh NIP transgenic T cells in the spleen at day 8 after priming.

C. PD-L1 expression on splenic DCs from mice treated with either isotype control, anti-IFNAR, anti-IL-10R, anti-PD-L1 or anti-PD-L1 plus IL-10R antibody (dual block – DB). Antibody treatment started one day prior to SMARTA priming and continued on day 1, day 4 and day 7 post-priming. This data is from the same experiment shown in Figure 1A. Bar graph depicts the geometric mean fluorescent intensity (GMFI) of PD-L1 on NK1.1- Thy1.2- CD45+ CD11c+ DCs in the spleen at Day 8 after SMARTA priming.

D. Bar graphs indicate LCMV titers in mice following antibody treatments described in Suppl Figure 1C. Titers are from Day 29 LCMV infection. This data is from the same experiment shown in Figure 1A.

Data are representative of 2 independent experiments with 3 – 5 mice per group. * $p < 0.05$.

Supplemental Figure 2 - Related to Figure 3. T-bet and Bcl6 expression in virus-specific CD4 T cells at the onset of acute and persistent LCMV infection with and without anti-PD-L1 / IL-10R blockade.

Virus-specific CD4 SMARTA T cells were transferred into mice 1 day prior to acute LCMV-Arm or persistent LCMV-Cl13 infection. LCMV-Cl13 infected mice were CD8 depleted prior to infection to avoid the mortality associated with anti-PD-L1 blockade at the onset of persistent viral infection. Mice were treated with isotype control or anti-PD-L1 and anti-IL-10R (Dual Block; DB) antibodies on day 0, 2, 5 and 8 of infection. Flow plots depict the frequency and bar graphs the number of T-bet^{hi} and Bcl6^{hi} CD4 SMARTA T cells on day 9 after infection. Data are representative of 2 independent experiments with 4 mice per group. * $p < 0.05$.

Supplemental Figure 3 - Related to Figure 4. Characterization of virus-specific CD4 SMARTA T cells immediately following 5 day *in vitro* Th1 polarization (pre-transfer) and at days 9 and 40 following priming.

A. The phenotype of Th1 polarized CD4 SMARTA T cells was assessed prior to transfer into LCMV-Cl13 infected mice. Histogram displays T-bet (red) and isotype (black) staining in Th1 polarized cells. Flow plots depict Th1 (SLAM^{hi} CXCR5^{lo}) and Tfh (SLAM^{lo} CXCR5^{hi}) CD4 SMARTA populations in Th1 polarized (Th1 Pol) cells and virus-specific CD4 SMARTA T cells from a day 5 LCMV-Armstrong infected mouse (positive control for Tfh staining), as well as IFNγ and TNFα production in unstimulated or PMA and ionomycin stimulated Th1 polarized cells.

B-D. Virus-specific CD4 SMARTA T cells were primed 21 days after LCMV-Cl13 infection (late primed; LP) or *in vitro* under Th1 polarizing conditions for 5 days (Th1 Pol) and injected into mice at day 26 post LCMV-Cl13 infection.

B. Flow plots show PSGL1 and Ly6C expression on late primed (LP) and Th1 polarized CD4 SMARTA T cells at day 9 and day 40 after SMARTA priming. Tfh are Ly6C negative and PSGL1 negative.

C. Histograms depict T-bet, Bcl6 and PD-1 expression in late primed (black) and Th1 polarized (red) SMARTA CD4 T cells 9 days after priming (4 days after transfer into LCMV-Cl13 infected mice).

D. Flow plots represent the proportion of Bcl6 positive cells gated from either Th1 ($SLAM^{hi}$ $CXCR5^{lo}$) or Tfh ($SLAM^{lo}$ $CXCR5^{hi}$) populations from *in vivo* late primed and Th1 polarized SMARTA CD4 T cells at day 40 after priming.

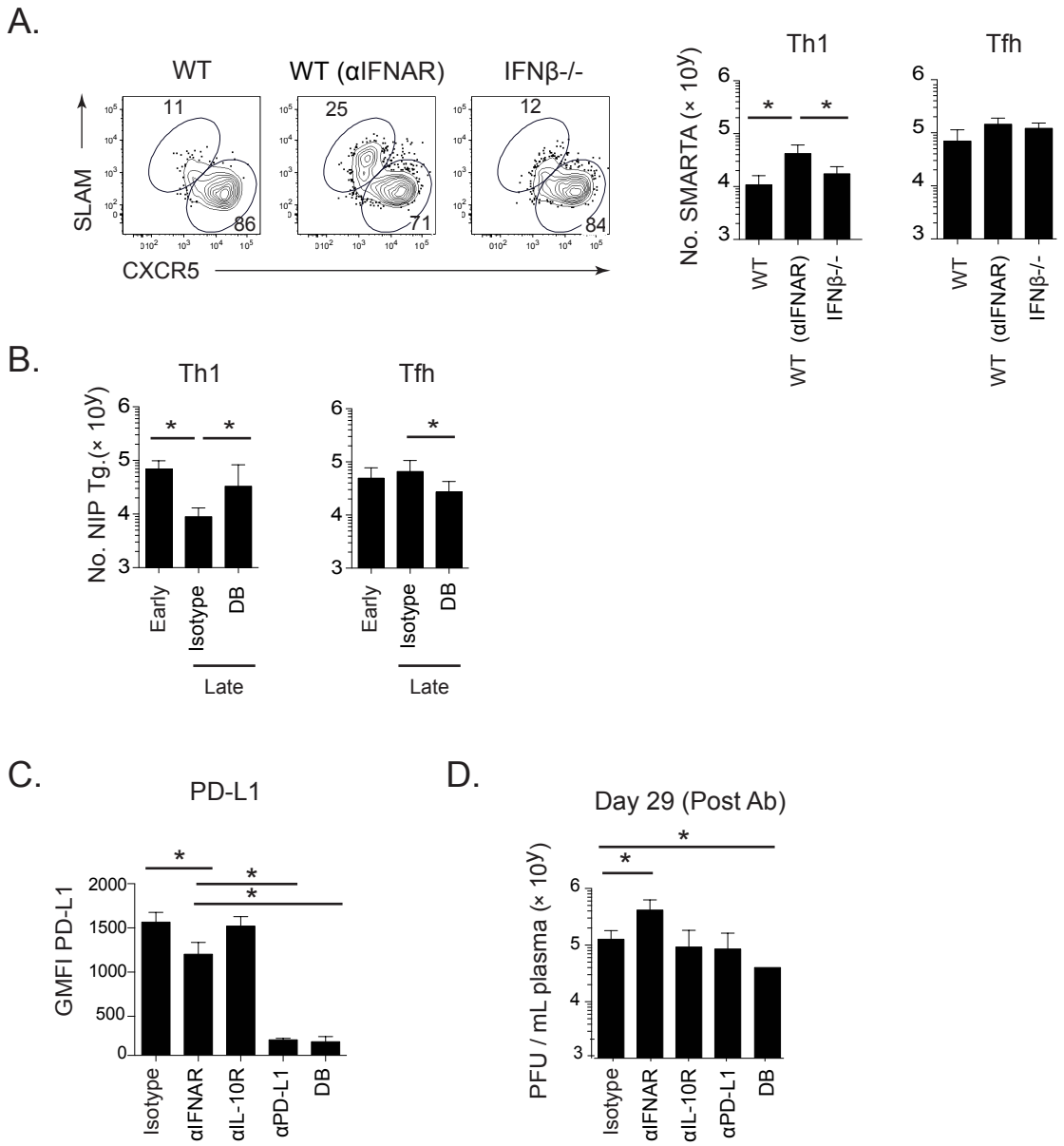
E. Bar graph represents the geometric mean fluourescent intensity (GMFI), and the histogram depicts PD-1 expression on late primed (black) and Th1 polarized (red) SMARTA CD4 T cells 40 days after priming.

Data are representative of 3 independent experiments with 3 - 5 mice per group. * $p < 0.05$.

Supplemental Figure 4 - Related to Figure 5. Th1 polarized virus-specific CD4 T cells enhance LCMV-specific CD8 T cells and antibody responses in CD4 depleted mice.

A-C. Mice were CD4 depleted prior to LCMV-Cl13 infection to assess effects of transferred CD4 SMARTA T cells in the absence of accessory help from pre-established LCMV-specific CD4 T cells or effects due to changes in virus titer. Naïve virus-specific CD4 SMARTA T cells were primed 21 days after LCMV-Cl13 infection (late primed; LP) or primed *in vitro* under Th1 polarizing conditions for 5 days (Th1 Pol) and injected into mice at day 26 post LCMV-Cl13 infection. As a control, a group of LCMV-Cl13 infected mice did not receive SMARTA cells (No cells). Analysis was performed in the spleen 40 days after SMARTA transfer (day 61 post LCMV infection). (**A**) Plasma virus titers (**B**) LCMV D^b/GP_{33-41} tetramer positive CD8 T cells and IFNγ producing CD8 T cells following *ex vivo* peptide restimulation with Gp33 peptide and (**C**) total plasma IgG in mice that received either no cells (gray), naïve late primed SMARTA cells (black), or Th1 polarized SMARTA cells (red)**.**

Data are representative of 2 independent experiments with 3 - 5 mice per group. * $p < 0.05$.



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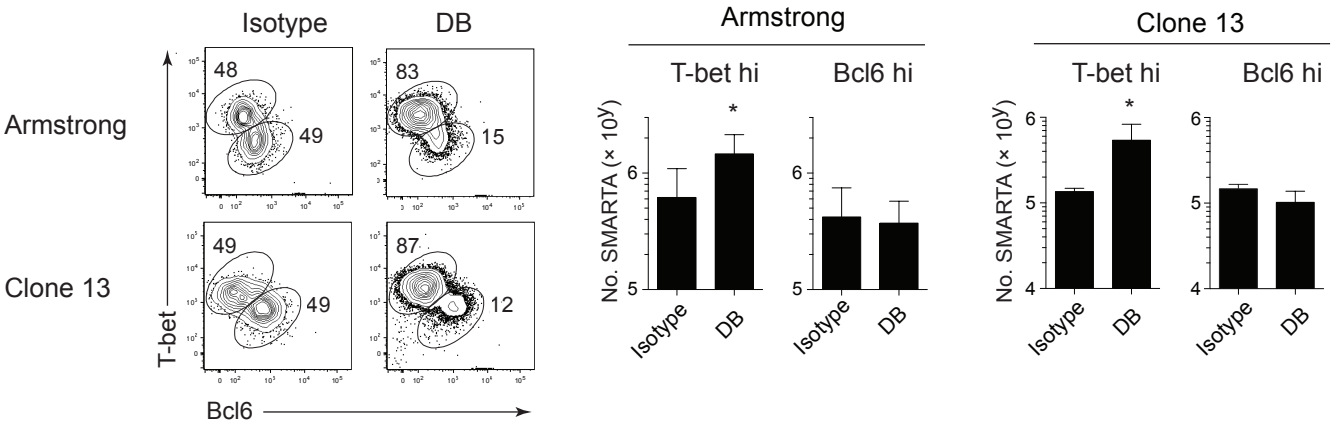

Figure S2. Related to Figure 3. T-bet and Bcl6 expression in virus-specific CD4 T cells at the onset of acute and persistent LCMV infection with and without anti-PD-L1 / IL-10R blockade.

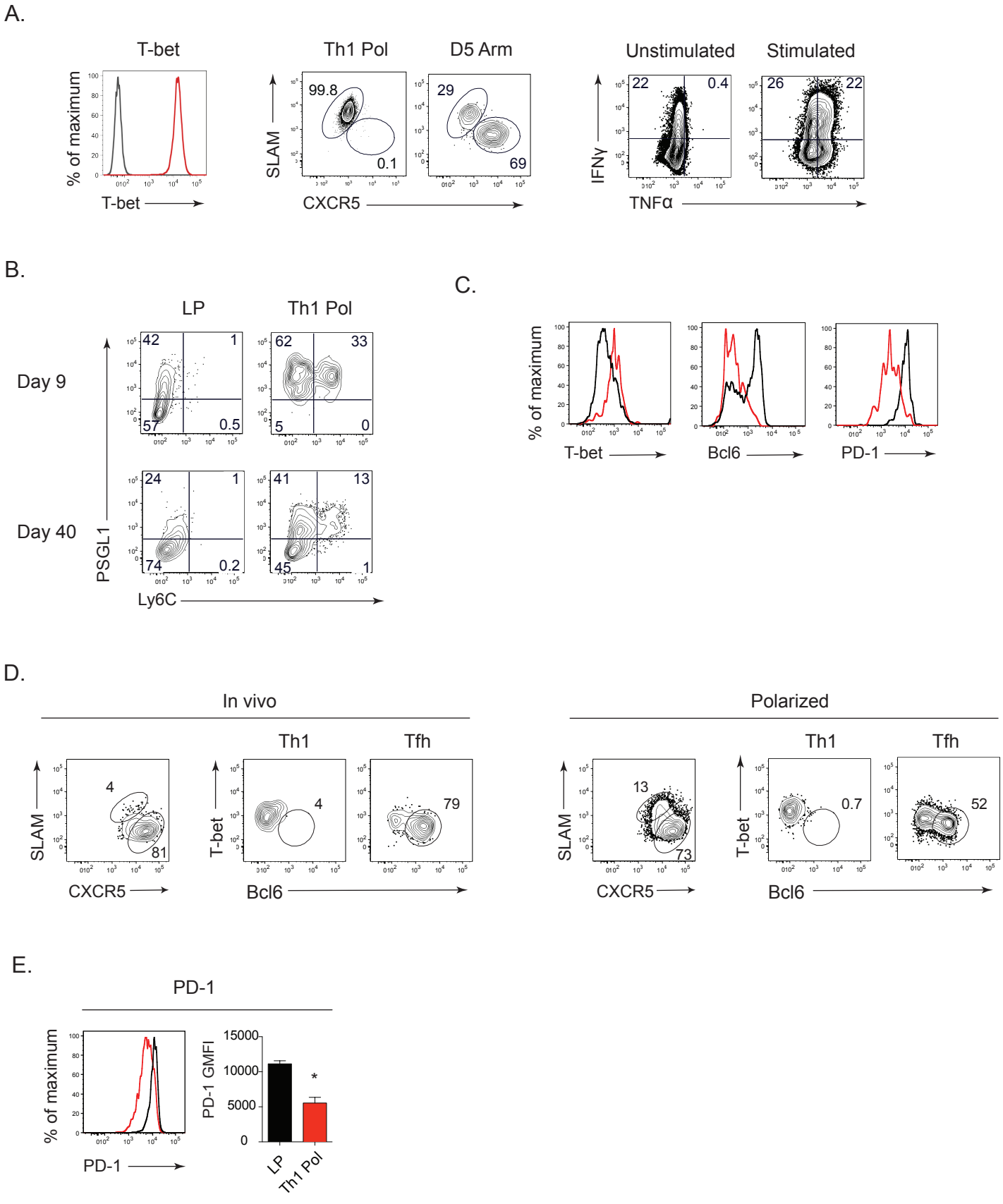

Figure S3. Related to Figure 4. Characterization of virus-specific CD4 SMARTA T cells pre-transfer immediately following 5 day in vitro Th1 polarization and at days 9 and 40 following priming.

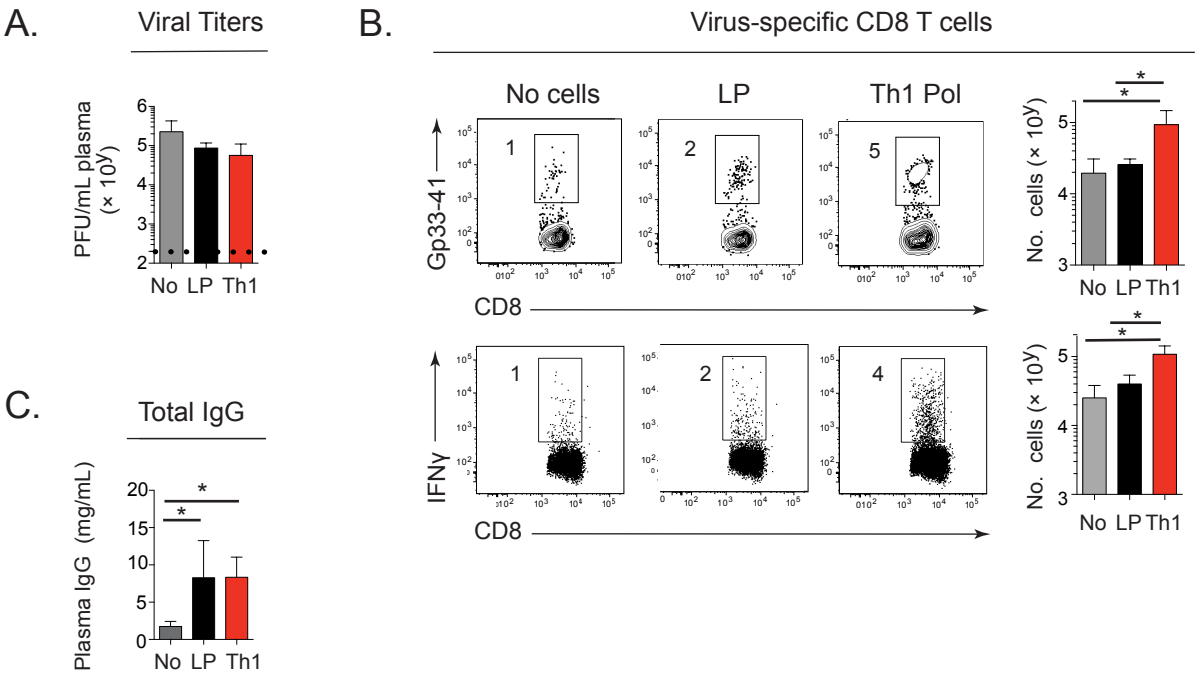

Figure S4. Related to Figure 5. Th1 polarized virus-specific CD4 T cells enhance LCMV-specific CD8 T cells and antibody responses in CD4 depleted mice.