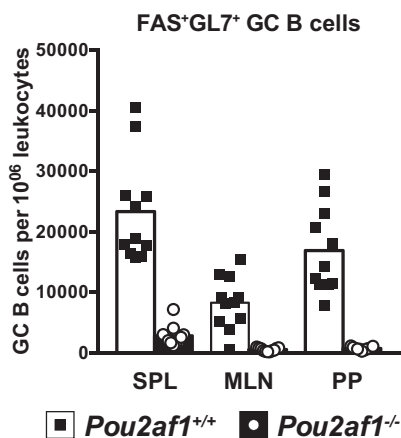
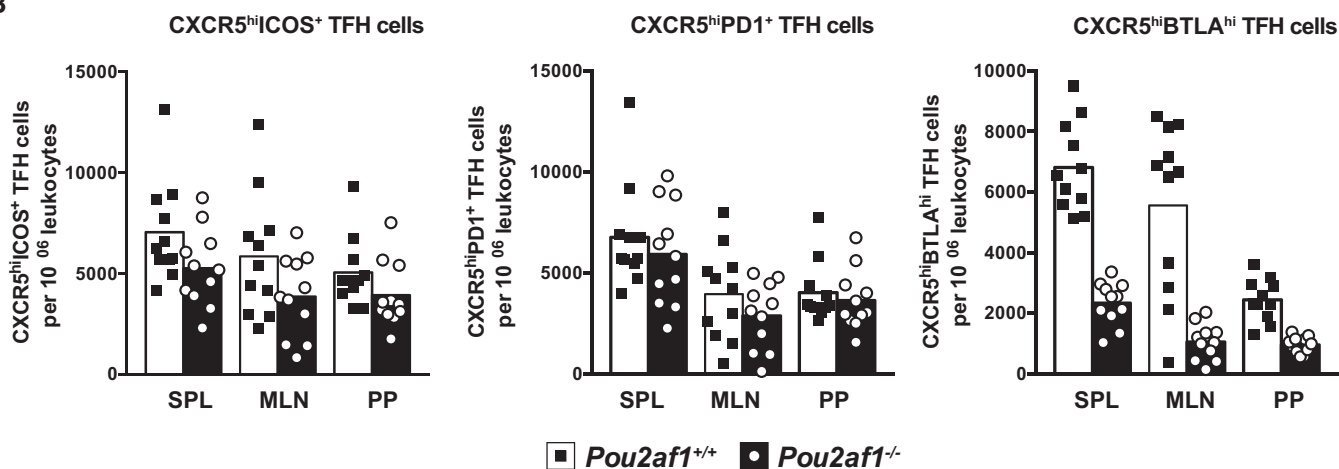


Expanded View Figures

A



B



C

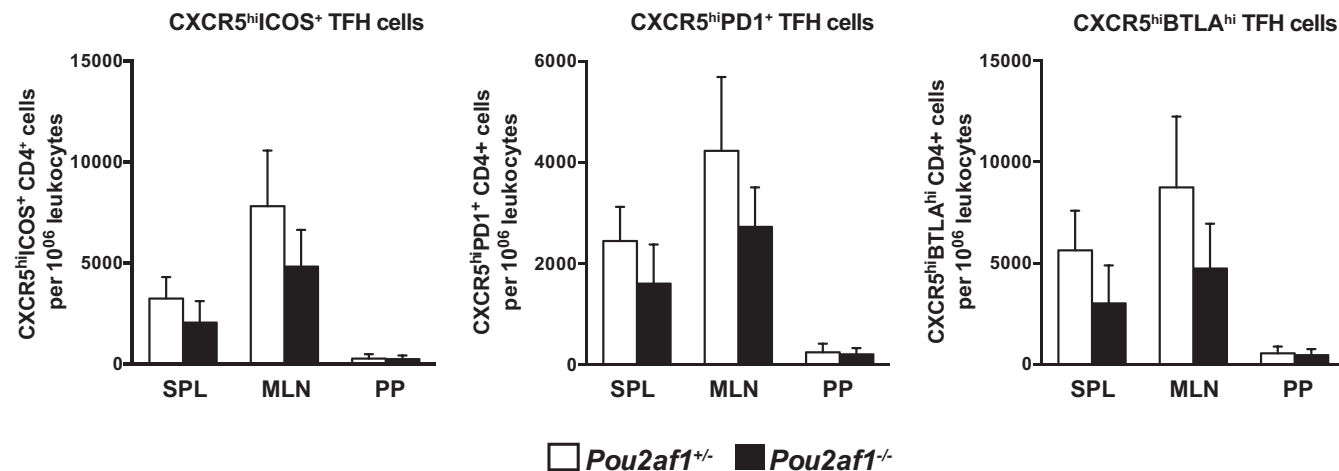
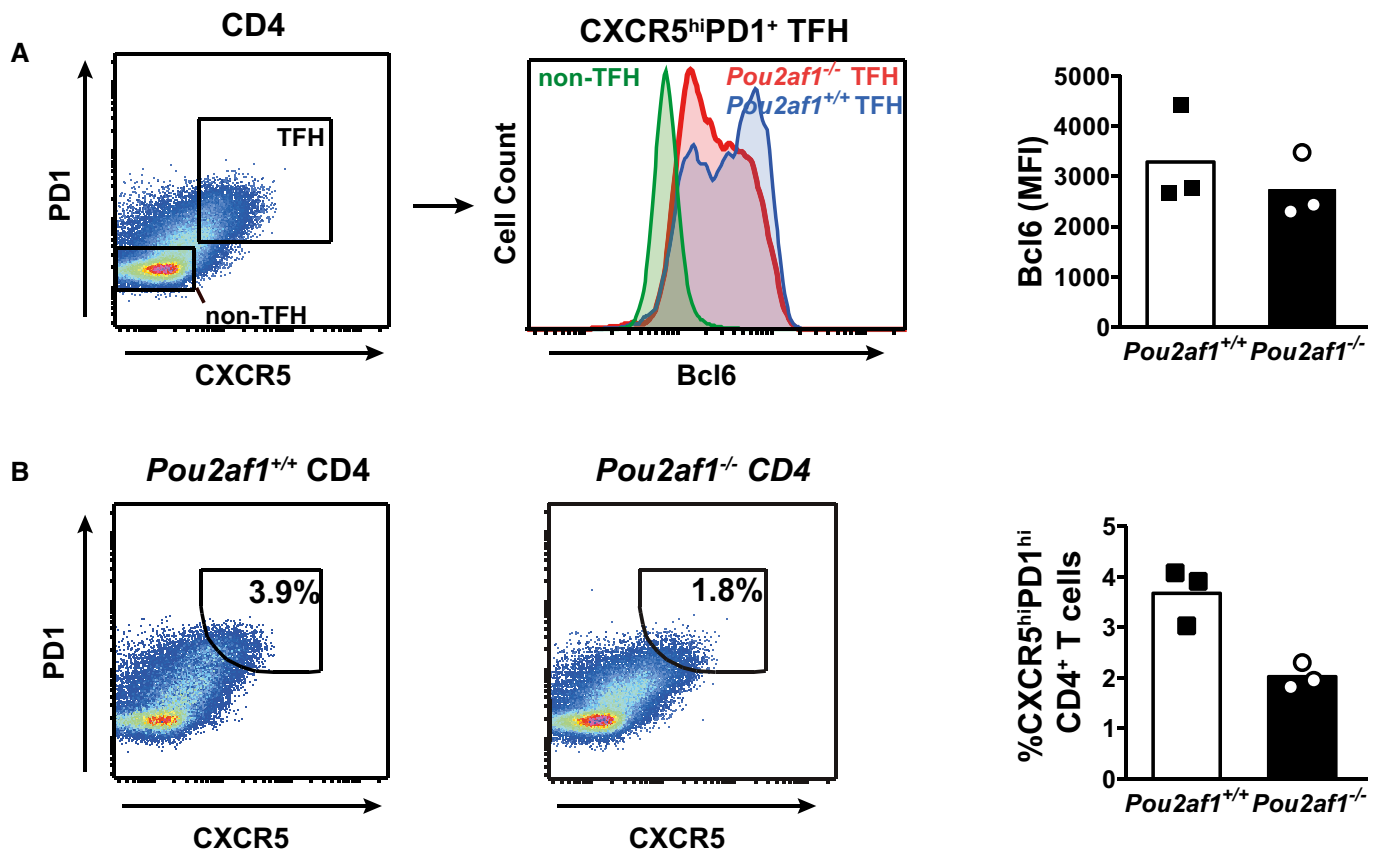


Figure EV1. T cell-intrinsic defect in the development of *Pou2af1*^{-/-} Tfh cells: cell numbers.

- A, B GC B-cell and Tfh cell development in the spleen (SPL), mesenteric lymph nodes (MLN), and Peyer's patches (PP) of *Pou2af1*^{+/-}:*Pou2af1*^{-/-} mixed bone marrow chimeras 7 days after immunization with SRBC. (A) Number of *Pou2af1*^{+/-} and *Pou2af1*^{-/-} B220⁺FAS⁺GL7⁺ GC B cells per million leukocytes. (B) Number of *Pou2af1*^{+/-} and *Pou2af1*^{-/-} Tfh cells—identified by the coexpression of CXCR5 and ICOS (CXCR5^{hi}ICOS⁺), CXCR5 and PD1 (CXCR5^{hi}PD1⁺), or CXCR5 and BTLA (CXCR5^{hi}BTLA^{hi}) on CD4⁺ T cells—per million leukocytes. The graphs show combined data from two independent experiments (*n* = 11) and correspond to the data shown in Fig 4B and C, respectively.
- C Tfh cell development in *CD4*^{-/-} mice reconstituted with *Pou2af1*^{+/-} or *Pou2af1*^{-/-} CD4⁺CD62L⁺OTII⁺ T cells. Mice were analyzed 7 days after immunization with OVA in alum for the presence of CXCR5^{hi}ICOS⁺, CXCR5^{hi}PD1⁺, and CXCR5^{hi}BTLA^{hi} Tfh cells. Bar graphs showing the cell number per million leukocytes correspond to the data shown in Fig 4D. Data are derived from two independent experiments with eight animals per group (mean ± SD).

**Figure EV2. *Pou2af1*^{-/-} PD1⁺ Tfh cells have a reduced capacity to develop into CXCR5^{hi}PD1^{hi} GC-Tfh cells.**

- A *Pou2af1*^{-/-} PD1⁺ Tfh cells express lower levels of Bcl6 than their wild-type counterparts in *Pou2af1*^{-/-}:*Pou2af1*^{+/-} mixed bone marrow chimeras. Bcl6 expression in PD1⁺CXCR5⁺CD4⁺ T cells was measured by flow cytometry on day 7 after immunization of mice with SRBCs (MFI: mean fluorescence intensity).
- B *Pou2af1*^{-/-} CD4⁺ T cells show a defect in the formation of GC-Tfh cells. Frequencies of CXCR5^{hi}PD1^{hi} GC-Tfh cells were lower for *Pou2af1*^{-/-} Tfh cells compared to wild-type Tfh cells in mixed bone marrow chimeras on day 7 after immunization with SRBCs.