

Supporting Information

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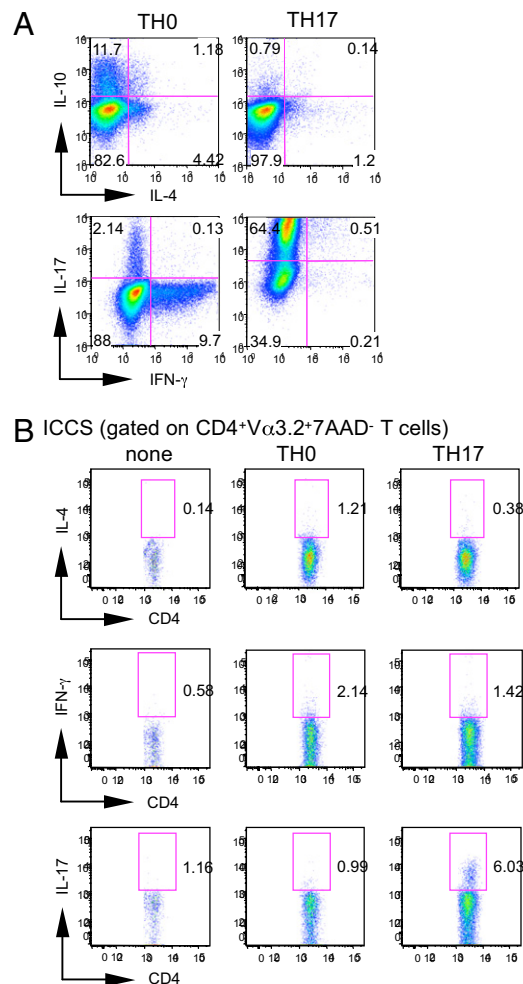


Fig. S1. (A) CD4⁺ T cells were stimulated with anti-CD3 (2.5 μg/mL) and IL-6 (25 ng/mL) plus TGF-β (2.5 ng/mL) for T helper type 17 (TH17) cells or no cytokines for the TH0 condition in the presence of irradiated CD4-depleted antigen-presenting cells. On day 3, cells were stimulated with phorbol 12-myristate 13-acetate (PMA) (50 ng/mL) and ionomycin (1 μg/mL) in the presence of monensin (1 μl/mL), and cytokine production was analyzed by intracellular cytokine staining for IL-4 and IL-10 or IFN-γ and IL-17. The gate was set on CD4⁺ T cells. (B) WT B6 mice were immunized with 50 μg recombinant myelin oligodendrocyte glycoprotein emulsified in incomplete Freund's adjuvant s.c. in both flanks and received at the same time 5×10^6 differentiated 2D2 T-cell receptor transgenic TH0 or TH17 cells i.v. Lymph node cells were purified 7 d after transfer/immunization and stimulated for 4.5 h with PMA (50 ng/mL) and ionomycin (1 μg/mL) in the presence of monensin (1 μl/mL). Subsequently, cytokine production was determined by intracellular staining for IL-4, IFN-γ, and IL-17. Gate was set on Vα3.2⁺CD4⁺7AAD⁻ T cells.

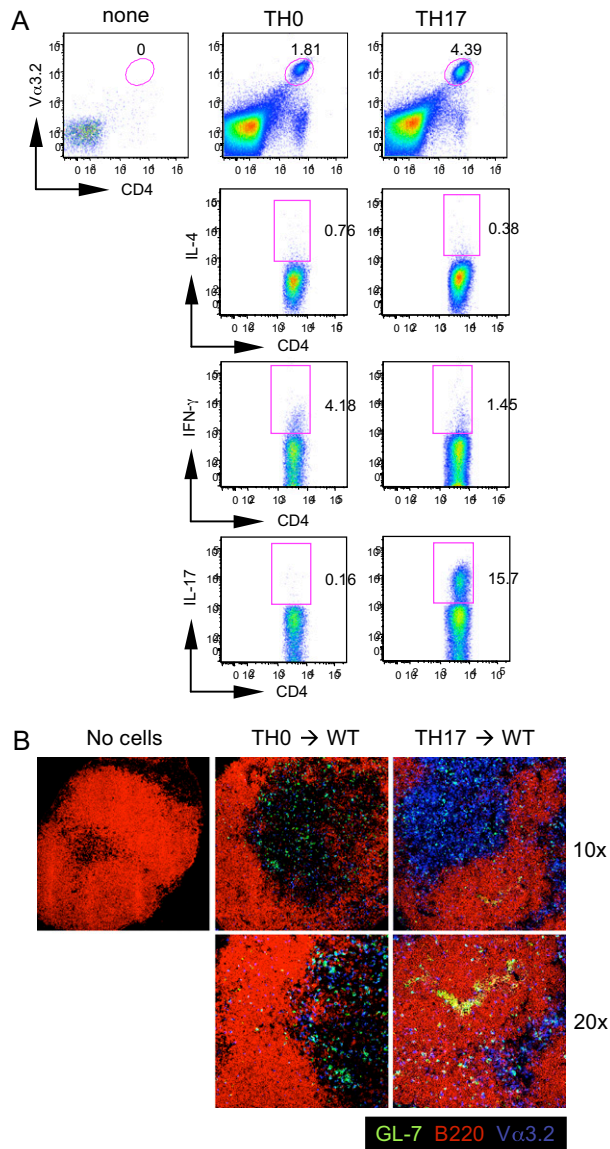


Fig. S3. T-cell receptor (TCR) α KO B6 mice were immunized with 50 μ g recombinant myelin oligodendrocyte glycoprotein (MOG) emulsified in incomplete Freund's adjuvant s.c. in both flanks and received at the same time 5×10^6 differentiated 2D2 TCR transgenic TH0 or TH17 cells intravenously. (A) On day 7, single-cell suspension of the draining lymph nodes was prepared, and cells were stimulated with phorbol 12-myristate 13-acetate (50 ng/mL) and ionomycin (1 μ g/mL) plus monensin (1 μ L/mL). Top row shows percentage of V α 3.2⁺CD4⁺ T cells in TH0 and TH17 recipient mice or control mice that did not receive any T cells. Lower rows show cytokine production (IL-4, IFN- γ , and IL-17) of V α 3.2⁺CD4⁺ T cells in TH0 and TH17 recipients. (B) On day 7, formation of germinal centers was analyzed by staining of lymph node sections for GL-7-FITC and B220-PE. Transferred T cells were detected with a V α 3.2 antibody. Lymph node sections were analyzed with a $\times 10$ objective (*Upper*) and $\times 20$ objective (*Lower*).