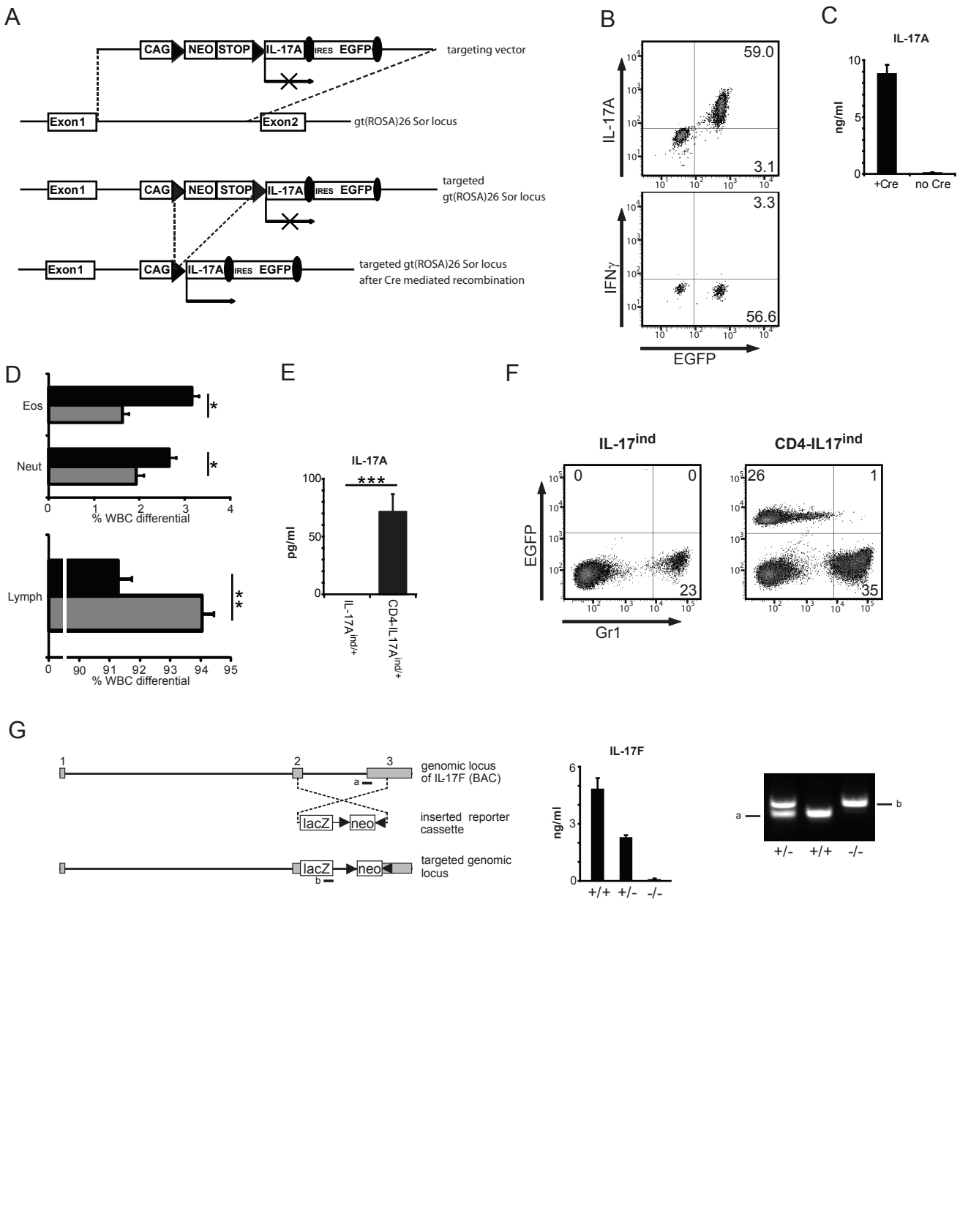
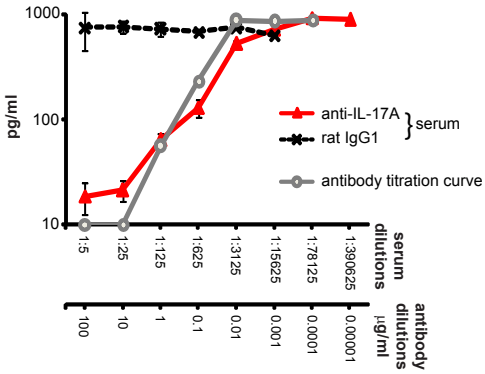
Supplemental Figure 1

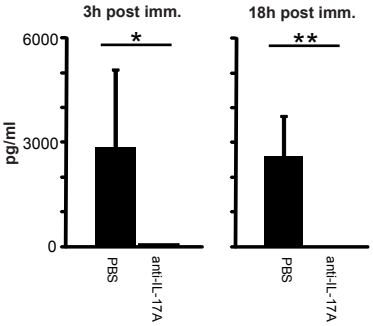


Supplemental Figure 2

A



B



Supplemental Figure 1 (A) Generation of the IL-17A^{ind} allele was carried out using homologous recombination in embryonic stem cells (C57BL/6-Bruce 4). Our conditional ‚knock-in' approach introduced the targeting construct into the endogenous gt(ROSA)26Sor locus. Upon Cre-mediated recombination, a lox-P-flanked transcriptional STOP cassette is excised 5' of an IL-17A cDNA insert and an IRES-EGFP element, allowing a dual expression of IL-17A and EGFP under the control of the chicken β-actin (CAG) promoter. (B) 1x10^6 whole lymph node cells from IL-17A$^{ind/+}$ mice were cultured for 4 h with or without (data not shown) Tat-Cre protein to induce IL-17A and EGFP expression in the presence of anti-CD3, anti-CD28 and IL-2. After 36 h, cells were briefly cultured with Brefeldin-A and stained for CD4 and either IL-17A or IFNγ. Percentages of IL-17A$^+$EGFP$^+$ and IFN-γ$^+$EGFP$^+$ cells are given in the quadrants. No EGFP was detected in wild type mice also treated with Tat-Cre (data not shown). (C) LN cells were cultured as in B, and supernatants extracted after 36 h. IL-17A secretion was measured by ELISA. (D) Peripheral blood was extracted from IL-17A$^{ind/+}$ and CD4-IL17A$^{ind/+}$ mice and subjected to routine WBC differential. Significant increase in neutrophil and eosinophil counts, and a decrease in peripheral blood lymphocytes are shown. (CD4-IL17A$^{ind/+}$ n=10, IL-17A$^{ind/+}$ n=9). (E) Serum from IL-17A$^{ind/+}$ and CD4-IL17A$^{ind/+}$ mice was extracted at d14 after immunization with MOG/CFA. Shown are levels of IL-17A detectable in serum isolated from peripheral blood (n=5). Spleen cells from the same mice were stained for Gr1. Quadrant statistics are shown. (F) Spleen-derived single cell suspension were obtained from the indicated genotypes d14 after MOG/CFA immunization. Percentages of Gr1$^+$ cells are shown in the histograms. (G) IL-17F gene-targeting strategy for the generation of the IL-17F deficient mouse strain. A reporter gene/resistance cassette (*lacZ* gene and a Ub1-Em7 promoter driven neomycin resistance gene, *neo*, flanked by loxP sites, ▶) was introduced into *il17f* exon 2 and 3 of a BAC containing the IL-17F gene locus. 129S6SvEv/C57B/6F1 embryonic stem cells were

targeted with a BAC carrying the replaced *il17f* gene locus and were subsequently screened as described elsewhere (41). “a” and “b” represent amplicons for the wild type and targeted allele respectively. Complete loss of IL-17F in the IL-17^{null} mouse was confirmed by ELISA on supernatant of T_H17 polarized splenocytes.

Supplemental Figure 2 Specification of the IL-17A blocking capacity of *in vivo* applied antagonistic antibody (A) WT or IL-17A$^{-/-}$ mice (n=3) were immunized with MOG/CFA and pertussis toxin. On day 0 and 4 either 200mg of antagonistic anti-IL-17A antibody or isotype control antibody (rat IgG1) was administered intraperitonially and mice are bled on day 7. The recovered serum was titrated in an IL-17A specific ELISA onto 1000 pg of IL-17A protein standard. The serum was serially diluted as shown in the graph. To estimate the anti-IL-17A titer in the serum the neutralizing antibody was itself in parallel titrated onto the same amount of IL-17A protein standard. IL-17A levels were measured after a 1 hour pre-incubation of the serum/antibody with the IL-17A standard. The graph shows the pooled data of all anti-IL-17A and rat IgG1 sera with SEM indicated. (B) Two groups of CD4-IL17A$^{ind/+}$ littermate mice were immunized with MOG/CFA (n=3). After 10 days, either 100μg/mouse of anti-IL-17A or sham antibody was injected intravenously. Mice were subsequently bled and serum was isolated either 3h or 18h after the antibody treatment. IL-17A serum concentrations were measured by flow cytometric cytokine analysis. Statistical significances are indicated in the graph (one-tailed T test).

Supplemental Table 1 Homeostatic leukocyte composition in spleen and thymus of naïve CD4-IL17A$^{ind/+}$ and control mice.

SPLEEN	Percentage	
	CD4-IL17A$^{ind/+}$	IL-17A$^{ind/+}$
T_H cells (CD4$^+$)	18	19
CTL (CD8$^+$)	9	11
T_{Reg} cells (FoxP3$^+$)	2	2
NK cells (NK1.1$^+$)	5	5
B cells (B220$^+$)	59	60
Macrophages (CD11b$^+$)	6	5
DC (CD11c$^+$)	6	4

THYMUS	Percentage	
	CD4-IL17A$^{ind/+}$	IL-17A$^{ind/+}$
T_H (CD4$^+$)	15	16
CTL (CD8$^+$)	6	7
Double positive T_H cells	73	73

Supplemental Table 2 Homeostatic leukocyte composition in spleen and thymus of naïve IL-17F deficient and control mice.

SPLEEN	Percentage		
	+/+	+/-	-/-
T_H cells (CD4$^+$)	14	16	14
CTL (CD8$^+$)	20	21	23
T_{Reg} cells (FoxP3$^+$)	3	3	2
NK cells (CD3$^-$, NK1.1$^+$)	3	4	3
NK T cells (CD3$^+$, NK1.1$^+$)	2	3	2
B cells	38	38	37
Macrophages (CD11b$^+$)	4	4	4
DC (CD11c$^+$)	4	4	3

THYMUS	Percentage		
	+/+	+/-	-/-
T_H (CD4$^+$)	14	15	13
CTL (CD8$^+$)	5	6	4
Double positive T_H cells	78	78	80