Supplemental Information

Basophil-derived IL-6 regulates T_H17 cell differentiation and CD4 T cell immunity

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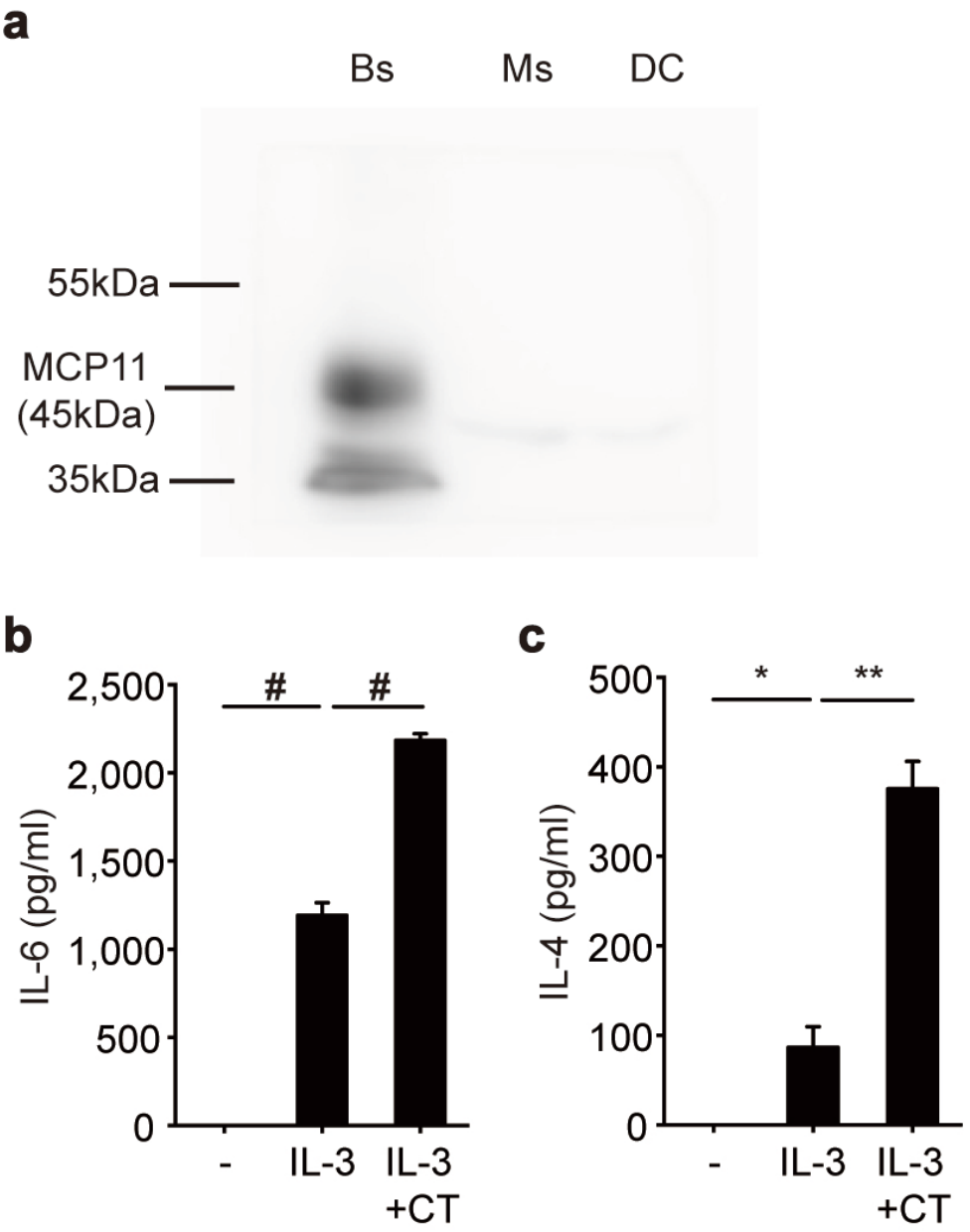
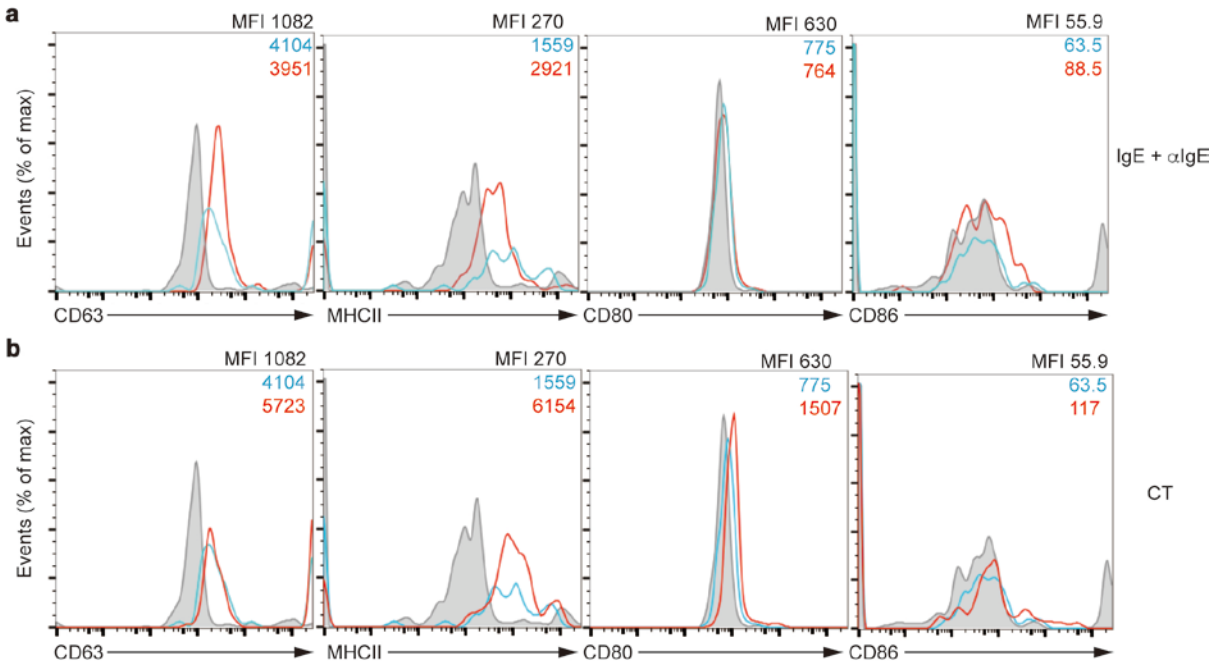


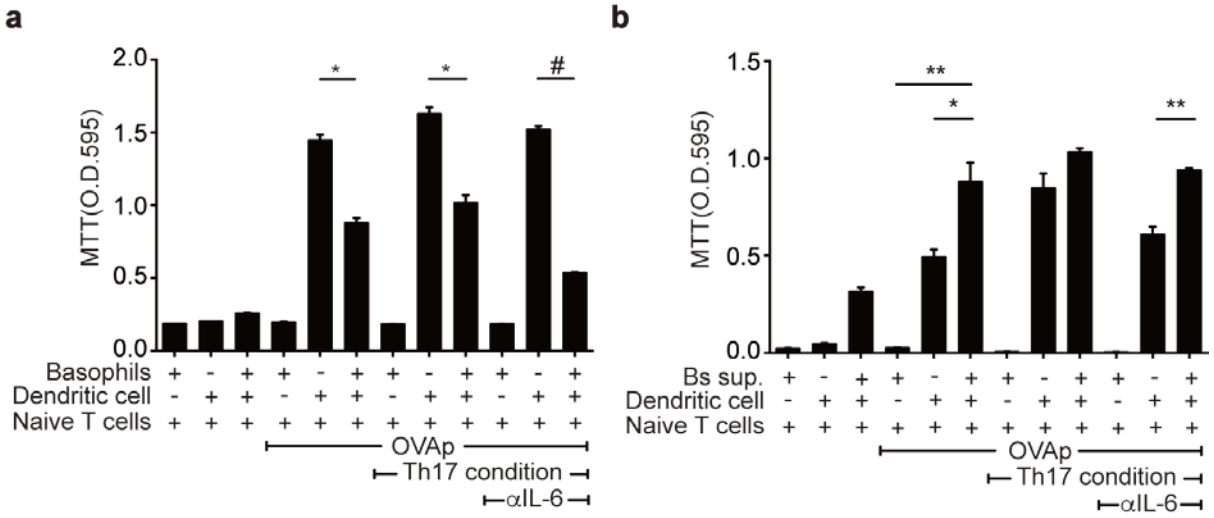
Supplemental Figure S1. IL-6 and IL-4 production by SPBs stimulated with IL-3 or IL-3 plus CT.

(a) Full-length blots of BMBs, BMMCs and BMDCs are presented for MCP-11 protein expression in Figure 1d. FACS-sorted YFP+ SPBs were stimulated with IL-3 or IL-3 plus CT for 24 hours. Secretion of IL-6 **(b)** and IL-4 **(c)** was quantified. Data are representative of three independent experiments (n=3).



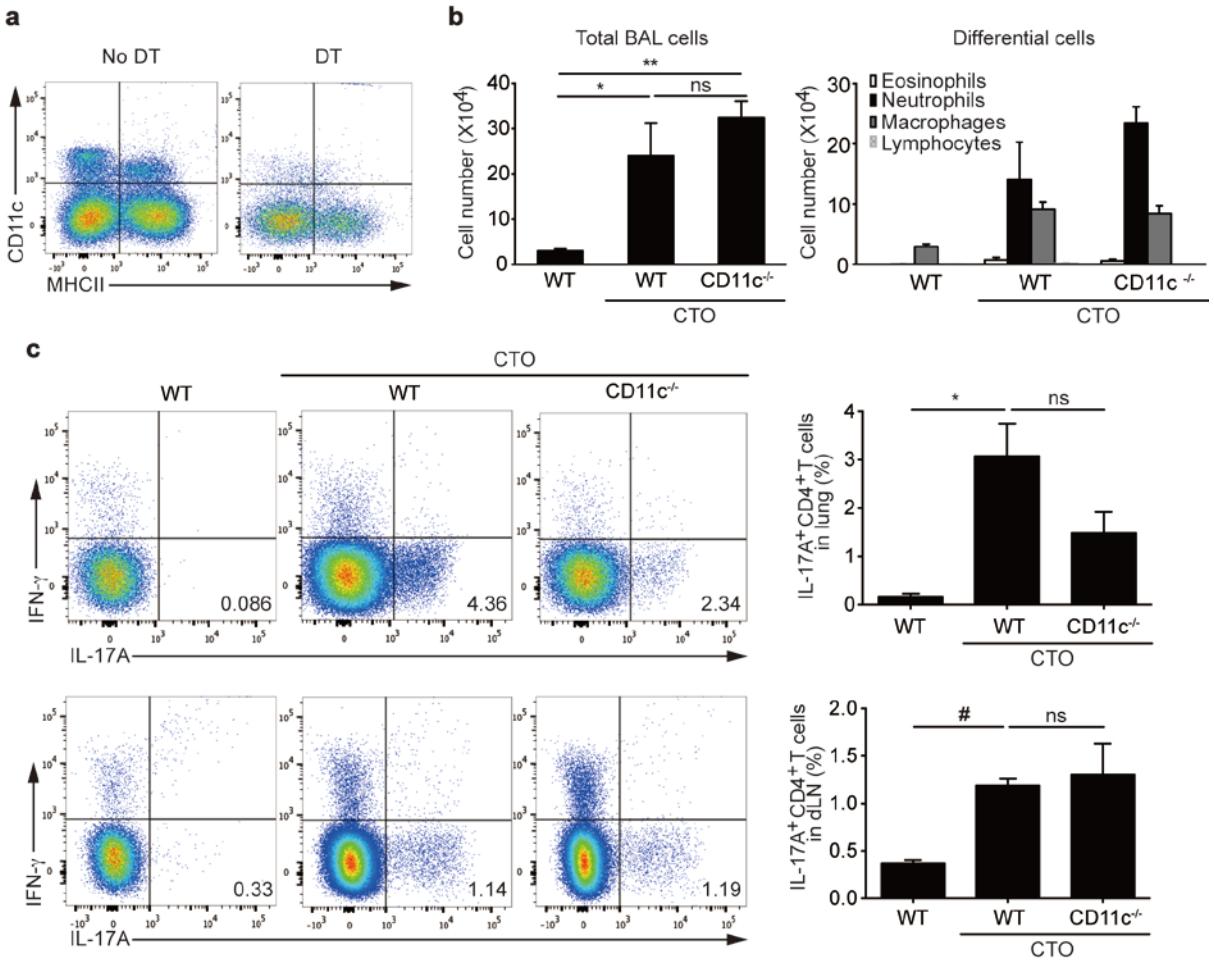
Supplemental Figure S2. Expression of cell surface markers on SPBs

Flow cytometric analysis of CD63, MHC class II, CD80, and CD86 expression on YFP^+ SPBs were examined after IgE crosslinking **(a)** or incubation with CT **(b)** (red line) or without stimulation (blue line) for 2 hours. Grey bars represent staining with isotype control antibody. Data are representative of three independent experiments.



Supplemental Figure S3. Cooperation of basophils and DCs to drive T_H17 cell proliferation

Proliferation of OT-II CD4 T cells incubated with DCs, basophils, both DCs and basophils **(a)** or supernatants of activated basophils **(b)** was assessed with MTT assay as in Figure 4. Data are representative of three independent experiments (n=3) and are presented as mean ± SEM.



Supplemental Figure S4. Normal induction of T_H17 responses in the absence of CD11c DCs

(a) $CD11c^+$ DCs were depleted in CD11c-DTR mice by DT treatment. DC depletion was confirmed by FACS analysis. **(b)** After administration of CTO, the numbers of total cells and inflammatory cells in BAL fluid were calculated in each group. **(c)** Intracellular cytokine staining of CD4 T cells from lungs and mediastinal LNs of CTO-immunized WT and DC depleted mice were assessed for the production of IL-17A and IFN-γ. Data are representative of three independent experiments (n=3) and are presented as mean ± SEM.