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Supplemental Information

Interleukin-17C Promotes Th17 Cell Responses

and Autoimmune Disease

via Interleukin-17 Receptor E

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Inventory

Five Supplemental Figures



Figure S1. Generation of $Il17c^{-/-}$ mice. A. Targeting strategy utilized for the insertion of a LacZ reporter cassette and the resulting disruption of the Il17c gene. Arrows indicate regions examined by PCR to verify cassette insertion and gene disruption. B. Genotyping of $Il17c^{+/+}$, $^{+/-}$, and $^{-/-}$ founder mice utilizing the PCR strategy described in A. C. Spleen and draining lymph nodes analysis of WT and $Il17c^{-/-}$ animals. Each plot is indicative of cells from one mouse where a total of 5 animals were examined for each staining condition. D. Splenocytes were stimulated with PMA and ionomycin and stained with IL-17A and IFN γ . The data shown are 3 different mice from each group, WT and $Il17c^{-/-}$ animals. Related to Figure 1.



Figure S2. Expression of IL-17R family in 293T cells. **A.** Expression of IL-17R family in 293T cells was confirmed by in vitro binding assay. After overexpression of control vector or vector containing IL-17Rs, 293T cells were incubated with hIg or IL-17 cytokine fusion proteins. Fusion protein binding was detected by anti-hIg conjugated with APC. **B.** Real-time RT-PCR of *Il17r* mRNA expression from 293T cells overexpressing IL-17R family. Related to Figure 2.



Figure S3. Intracellular cytokine staining of differentiated CD4 T cells. Naïve CD4 T cells were activated with anti-CD3 and anti-CD28 antibody under different polarizing conditions for 5 days and stained with cytokine antibodies or Foxp3. Related to Figure 3.



Figure S4. A. Real-time RT-PCR of *Il17re* mRNA expression. Naïve CD4 T cells were activated with anti-CD3 and anti-CD28 antibody under the indicated conditions with and without IL-17C for 4 days (*p<0.05). **B**. Binding of IL-17C-hIg to IL-17A positive cells isolated from CNS of EAE mice. CNS was collected from EAE mice at different disease scores. Mononuclear cells were collected after percoll and stained with hIg or IL-17C-hIg. Related to Figure 4.



Figure S5. Real-time RT-PCR analysis of transcription factors in CD4 T cells retrovirally transduced with IL-17RE or control vector and treated with IL-17C for 2 d. mRNA were isolated from sorted GFP^+ cells. Related to Figure 5.