

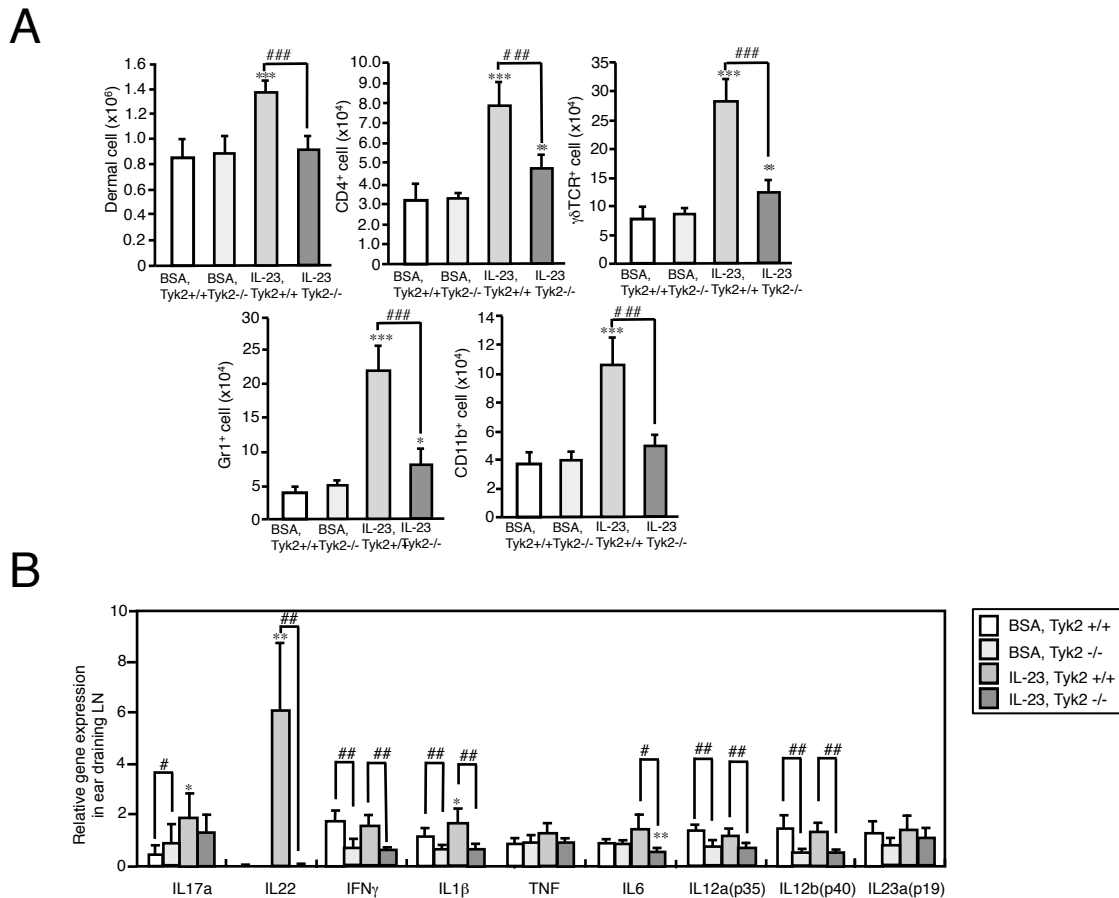


Title	Tyk2 is a therapeutic target for psoriasis-like skin inflammation
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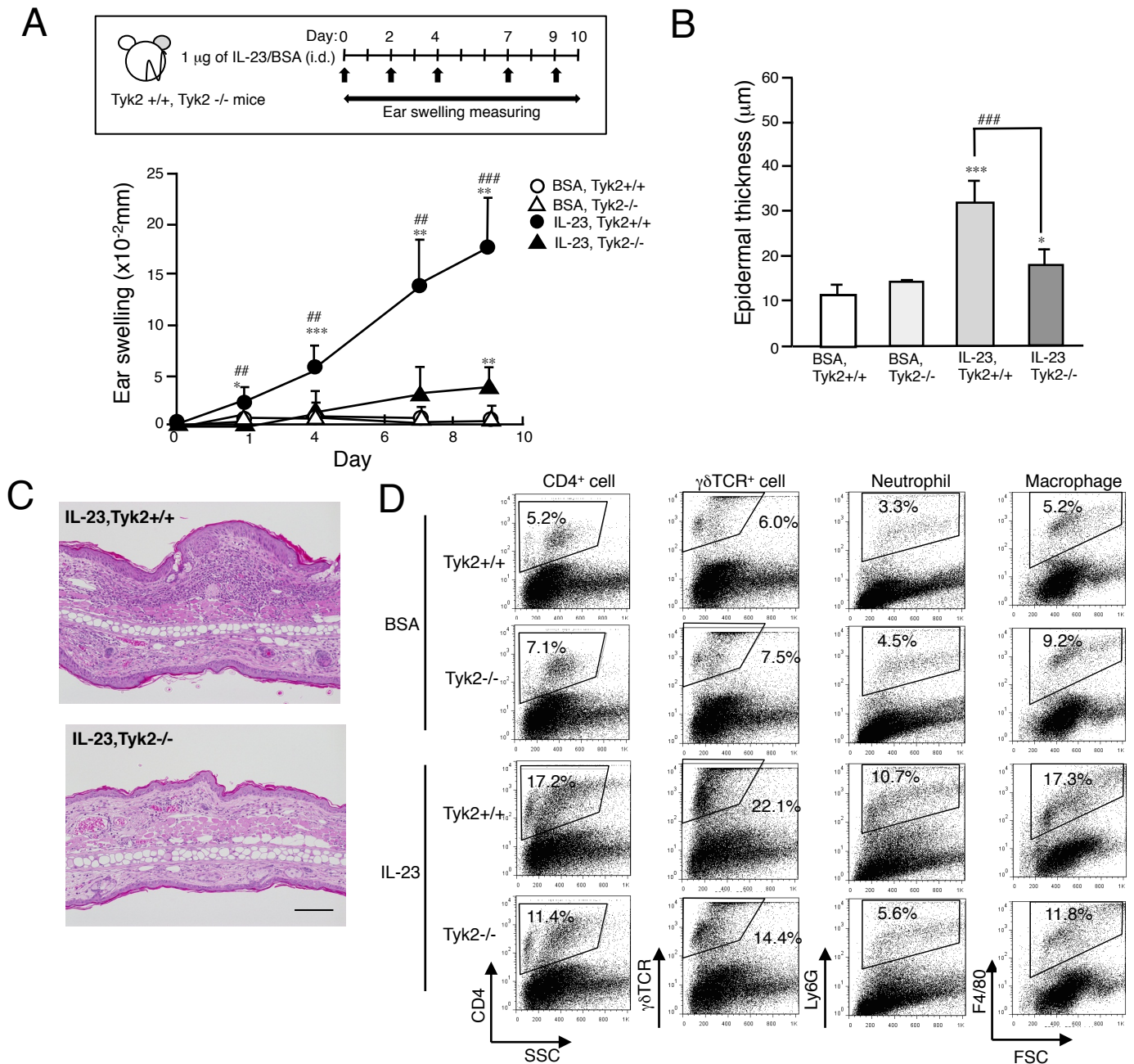
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Supplemental Fig. 1



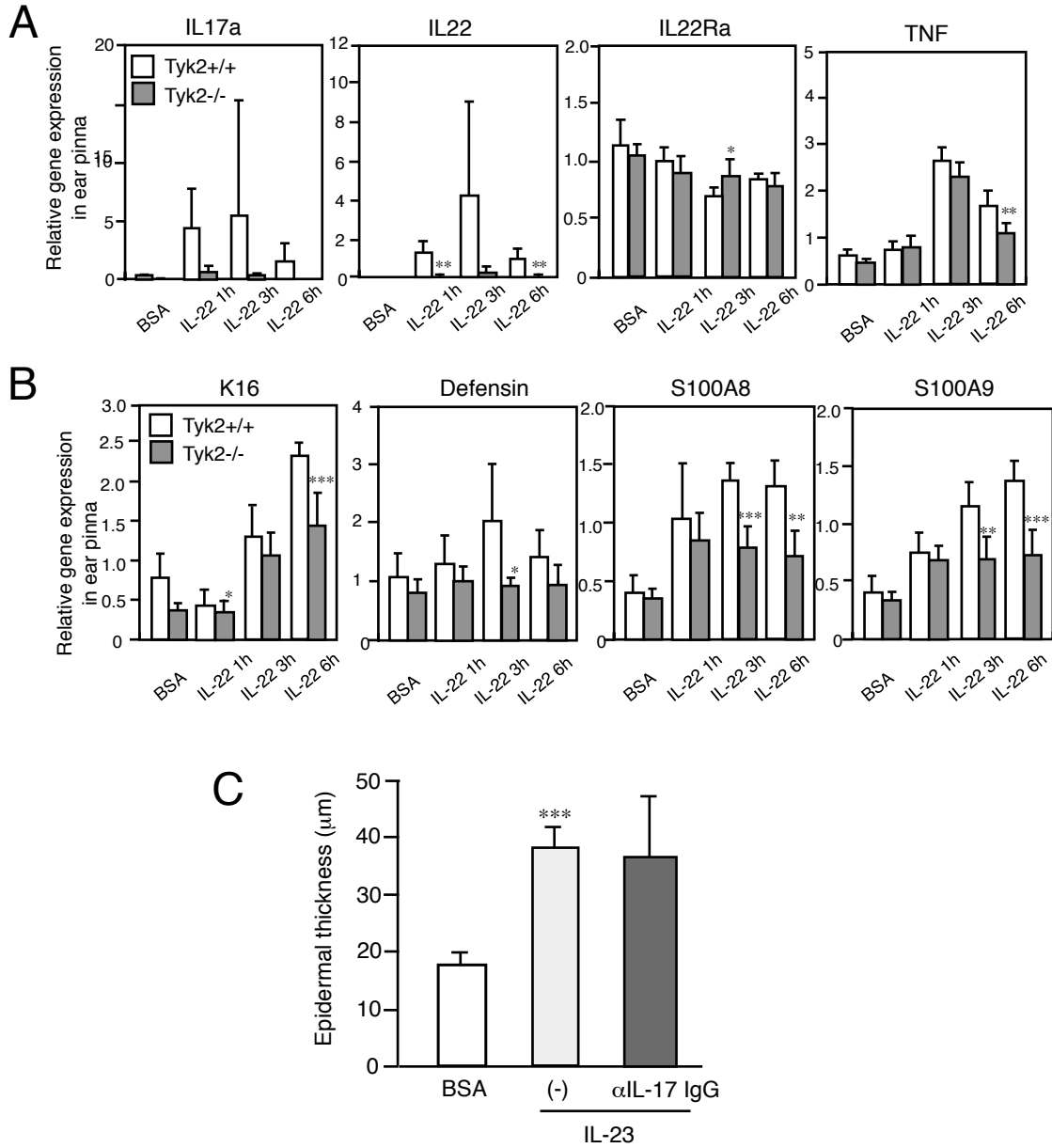
Supplementary Fig. 1. Dermal inflammatory cells are diminished by Tyk2 deficiency in IL-23-induced skin inflammation. Dermal cells were isolated from digested Tyk2^{+/+} and Tyk2^{-/-} ear skin on day 4, and subjected to flow cytometry. The absolute number of inflammatory cells from uniformly collected ear tissue by biopsy punch is shown. Data represents mean \pm SD (n=5). *p<0.01, ***p<0.001 compared with BSA-injected mice group; ###p<0.001 compared with Tyk2^{-/-} mice group. Similar results were obtained in two independent experiments. (B) Tyk2^{+/+} and Tyk2^{-/-} mice were intradermally injected with 1 μ g/ear of IL-23 or BSA for every day. mRNAs were prepared from the ear draining LN of Tyk2^{+/+} and Tyk2^{-/-} mice on day 4, and subjected to quantitative RT-PCR with the indicated primers. Gene expression level was normalized by GAPDH expression. Data represents mean \pm SD (n=5). *p<0.05, **p<0.01 compared with BSA-injected mice group; #p<0.05, ##p<0.01 compared with Tyk2^{-/-} mice group. Similar results were obtained in two independent experiments.

Supplemental Fig. 2



Supplementary Fig. 2. Involvement of Tyk2 in IL-23-induced prolonged skin inflammation. (A) The ear pinna of Tyk2^{+/+} and Tyk2^{-/-} mice was intradermally injected with 1 µg/ear of IL-23 or BSA on an intermissive schedule, 5-times in 10 days. Ear swelling was evaluated on the days as indicated. (B) Epidermal hyperplasia was evaluated by measuring the epidermal thickness by imaging analysis. Data represents mean ± SD (n=5). *p<0.05, **p<0.01, ***p<0.001 compared with BSA-injected mice group; ##p<0.01, ###p<0.001 compared with Tyk2^{-/-} mice group. (C) Representative H&E stained histological features of IL-23-treated ear pinna from Tyk2^{+/+} and Tyk2^{-/-} mice. Scale bar, 100 µm. (D) Flow cytometry analysis of the dermal cells isolated from pooled Tyk2^{+/+} and Tyk2^{-/-} ear skin by the intermissive injection. Surface markers of major inflammatory cells were analyzed with fluorescent-labeled specific antibodies. Similar results were obtained in two independent experiments.

Supplemental Fig. 3



Supplementary Fig. 3. Tyk2 directly regulates IL-22-induced skin inflammation. (A) IL-22-dependent inflammatory molecules expression in ear pinna. IL-22 was injected into the ear pinna, then the pinna was harvested from sacrificed Tyk2^{+/+} and Tyk2^{-/-} mice at 1, 3 and 6 h after the single injection. Data represents mean ± SD (n=5). *p<0.05, **p<0.01, ***p<0.001 compared with Tyk2^{+/+} mice group. (B) IL-22-dependent keratinocyte proliferation marker K16 and antimicrobial peptide expressions in the ear pinna. The ear pinna from Tyk2^{+/+} and Tyk2^{-/-} mice was harvested at 1, 3 and 6 h after the single IL-22 injection. Similar results were obtained in two independent experiments. (C) Epidermal hyperplasia is not depend on IL-17A in IL-23-induced skin inflammation. Anti-IL-17A neutralization antibody (0.1 mg/mouse) was intraperitoneally injected on day 0, then the ear pinna of Tyk2^{+/+} mice was intradermally injected with 1 μg/ear of IL-23 or BSA for 4 consecutive days. Epidermal thickness were evaluated on day 4. Data represents mean ± SD (n=5). ***p<0.001 compared with BSA. Similar results were obtained in two independent experiments.