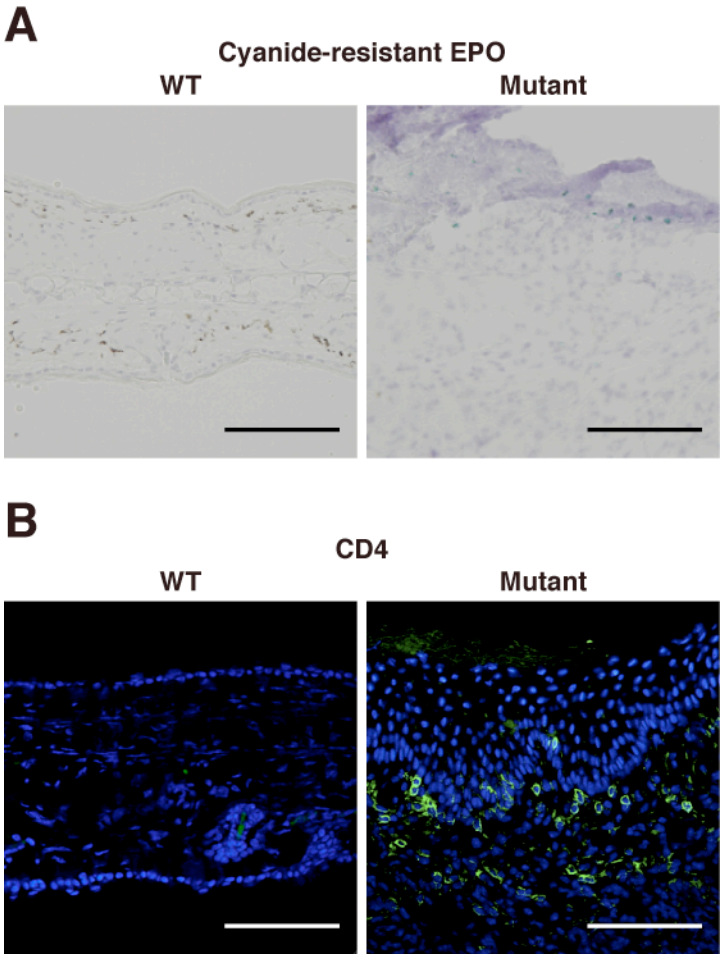
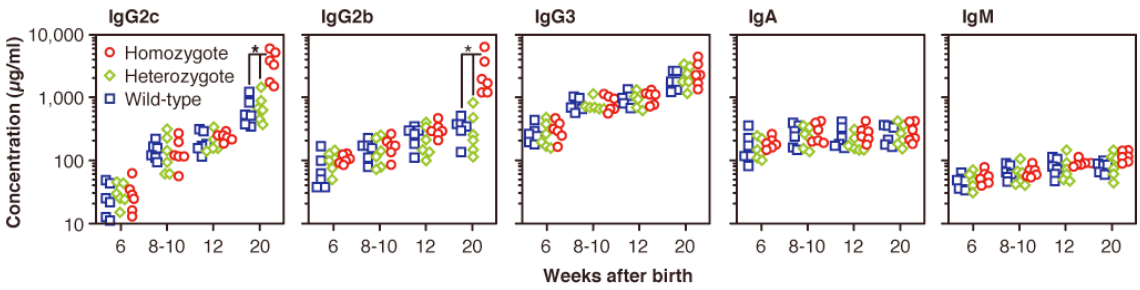


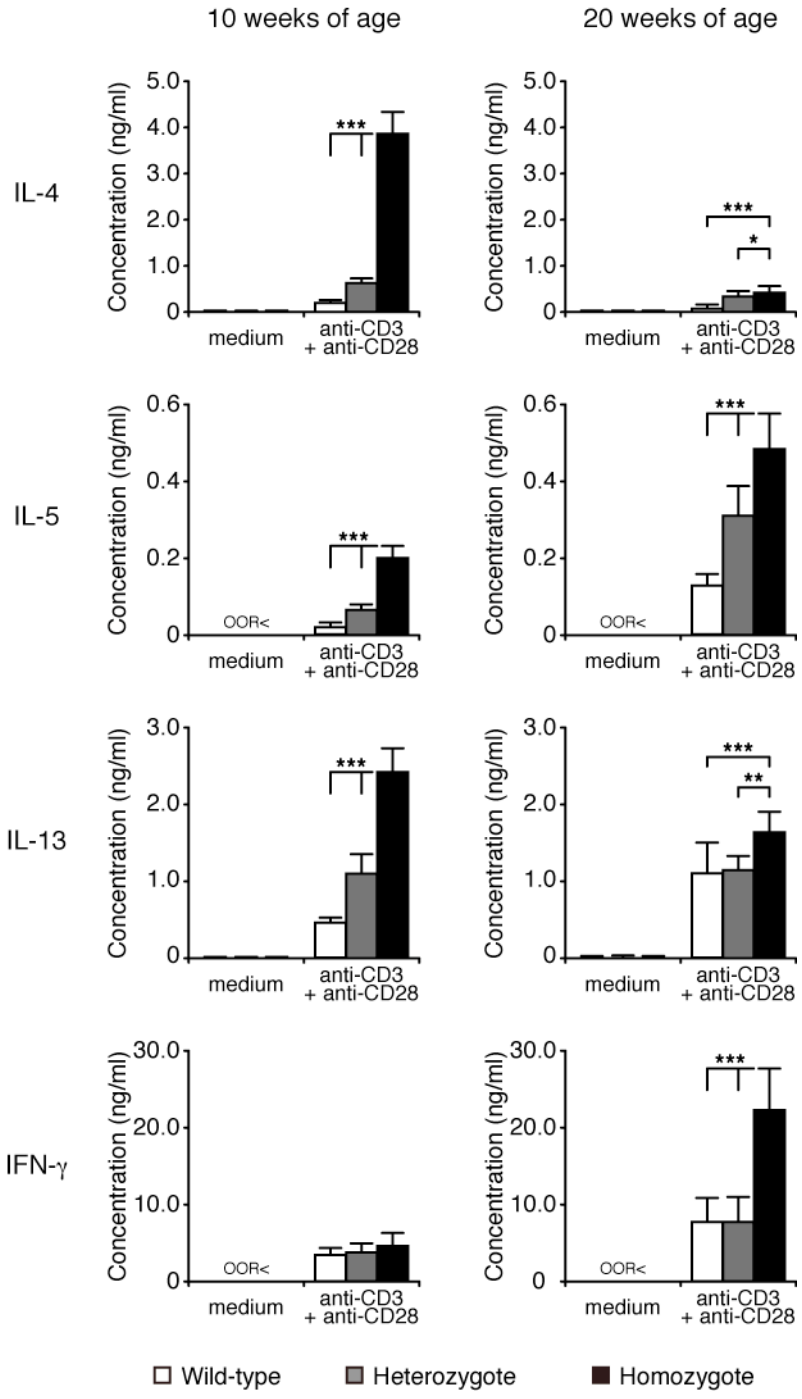
Supplemental Figure 1

Pathological findings in mutant mice. (**A** and **B**) Photomicrographs of histological sections of ear skin from mutant and WT mice (18 weeks of age) stained for eosinophils using a cyanide-resistant eosinophil peroxidase (EPO) assay (**A**) and for $CD4^+$ T cells by immunostaining with a CD4 antibody (**B**). Tissues were counterstained with hematoxylin (**A**) or DAPI (**B**). Scale bar, 100 μm.



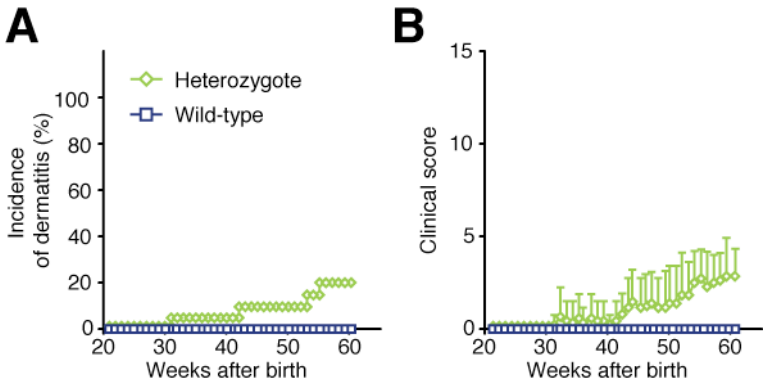
Supplemental Figure 2

Serum immunoglobulins in mutant mice. Longitudinal analyses of serum immunoglobulin levels in homozygous mutant, heterozygous mutant, and WT littermates. Each symbol represents an individual mouse and $*P < 0.05$, Bonferroni test following 1-way ANOVA.



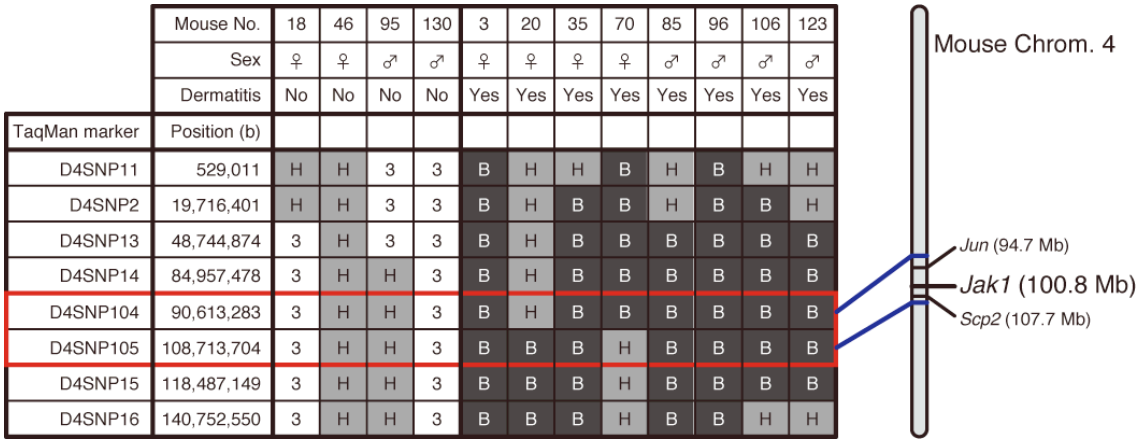
Supplemental Figure 3

CD4$^+$ T cell responses. CD4$^+$ T cells from spleen and LN of homozygotes, heterozygotes and WT mice at 10 weeks (early stage of dermatitis) and 20 weeks (chronic stage of dermatitis) of age were isolated, and stimulated with plate-coated CD3 (1.0 μg/ml) and CD28 (1.0 μg/ml) antibodies for 72 h, and cytokine production in culture supernatants was detected by suspension array system. Each bar represents the mean ± SD from triplicate cultures. OOR<, Out of range, below detectable level. *$P < 0.05$, **$P < 0.01$ and ***$P < 0.001$, Bonferroni test following 1-way ANOVA.



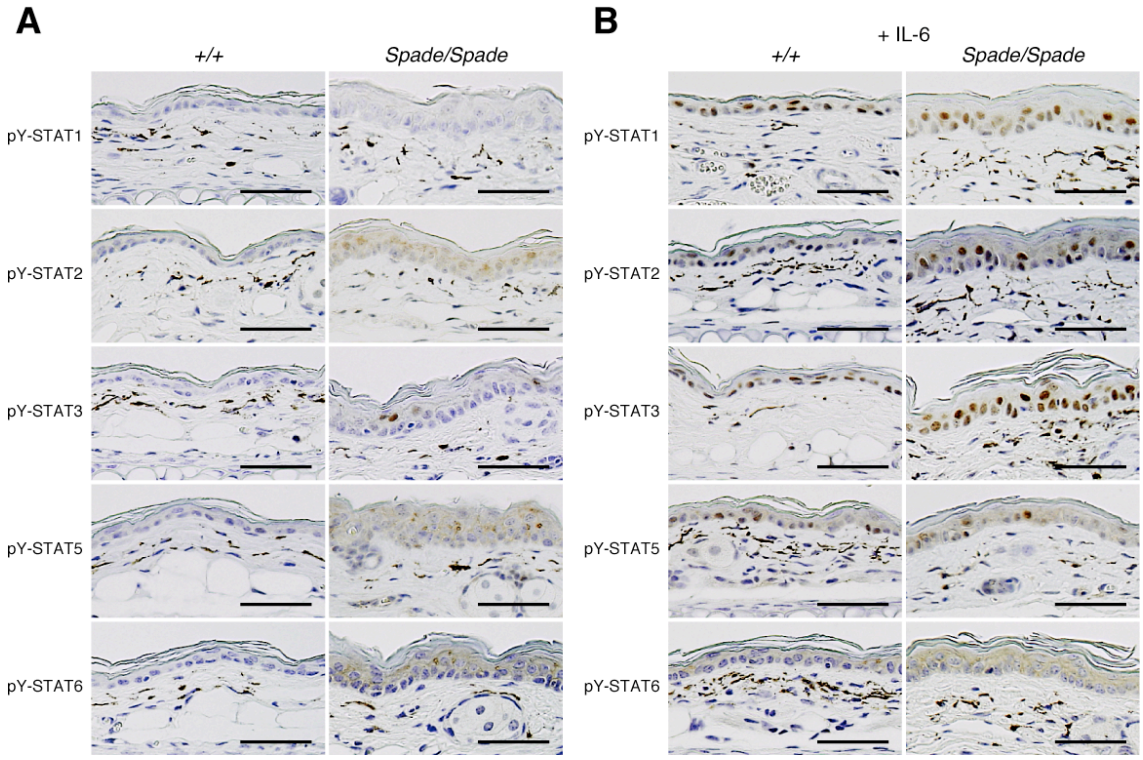
Supplemental Figure 4

Development of dermatitis in heterozygous mutant mice. (**A** and **B**) Longitudinal analyses of dermatitis incidence (**A**) and clinical scores (**B**) in heterozygous mutant mice. The cumulative incidence of dermatitis was determined from cohorts of 20 heterozygous and 50 WT mice, and data in **B** are mean ± SD.



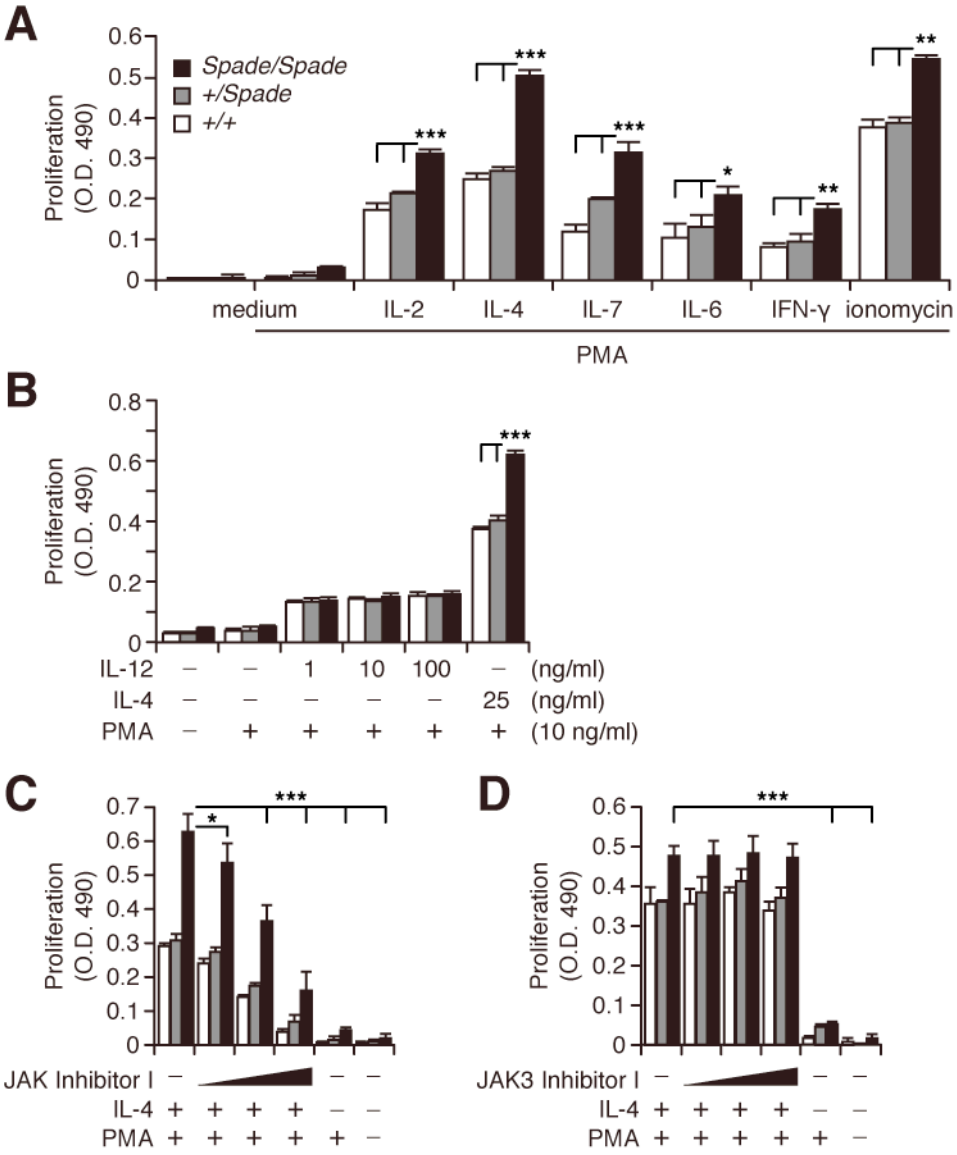
Supplemental Figure 5

Meiotic mapping of the *Spade* mutation. The dermatitis phenotype was mapped to the region from the SNP marker D4SNP104 (90.6 Mb) to D4SNP105 (108.7 Mb) on chromosome 4. Several candidate genes, including Jak1, have been mapped to this region. The genotype for each strain is represented as B (C57BL/6J homozygote), 3 (C3H/HeJ homozygote), or H (heterozygote).



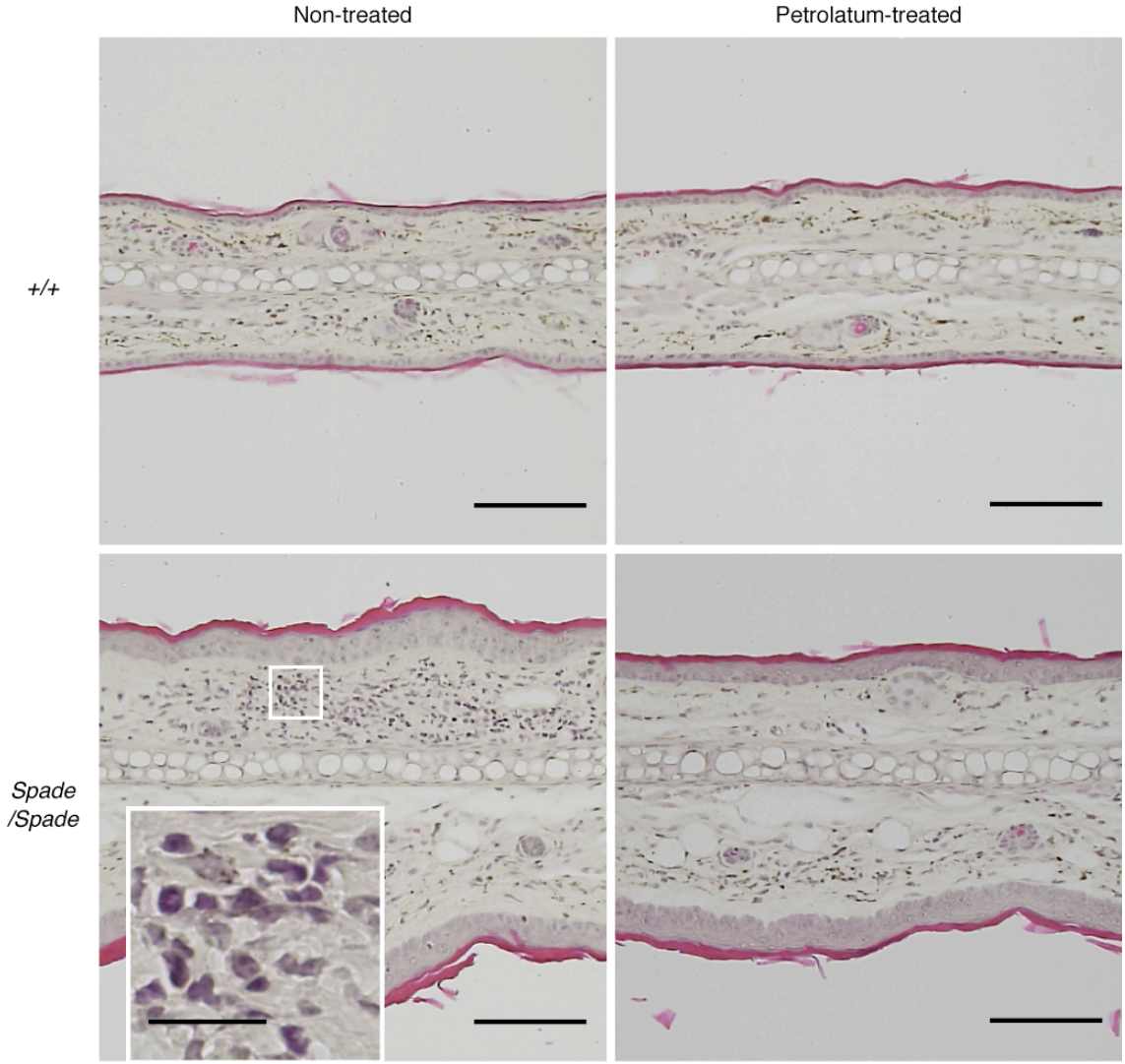
Supplemental Figure 6

Immunohistological analysis of Stat activation in *Spade/Spade* and WT mice. (**A**) Photomicrographs of ear skin histological sections from 6 weeks of age mutant and WT mice immunostained for pY-STAT1, 2, 3, 5 and 6. (**B**) As in **A**, except that the ear skin was harvested 30 min after the intraperitoneal injection of IL-6. Scale bar, 20 μm.



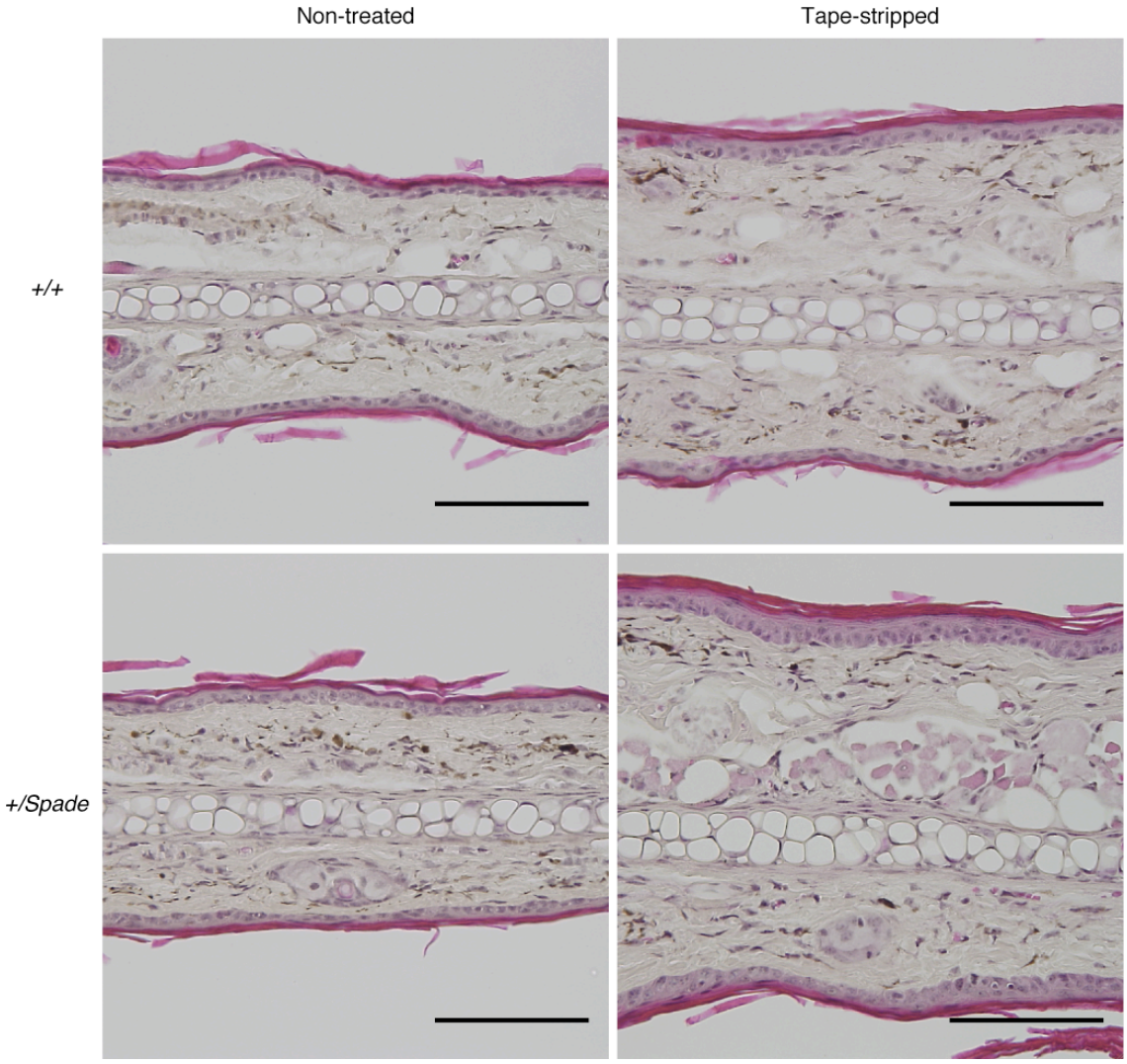
Supplemental Figure 7

Proliferative responses of mutant lymphoid cells. (**A** and **B**) Thymocytes from the *Spade/Spade*, *+/Spade* and WT littermates were cultured with or without cytokines or ionomycin in the presence of phorbol myristate acetate (PMA). Cell proliferation was measured 48 h later using a soluble tetrazolium/formazan assay. (**C** and **D**) Thymocytes were stimulated with or without IL-4 and PMA in the absence or presence of the various concentrations of a pan-Jak inhibitor (JAK inhibitor I, **C**) or a Jak3-specific inhibitor (JAK3 inhibitor I, **D**). Cell proliferation was measured as in **A**. The results represent the mean ± SD from triplicate cultures. **P* < 0.05, ***P* < 0.01 and ****P* < 0.001, Bonferroni test following 1-way ANOVA.



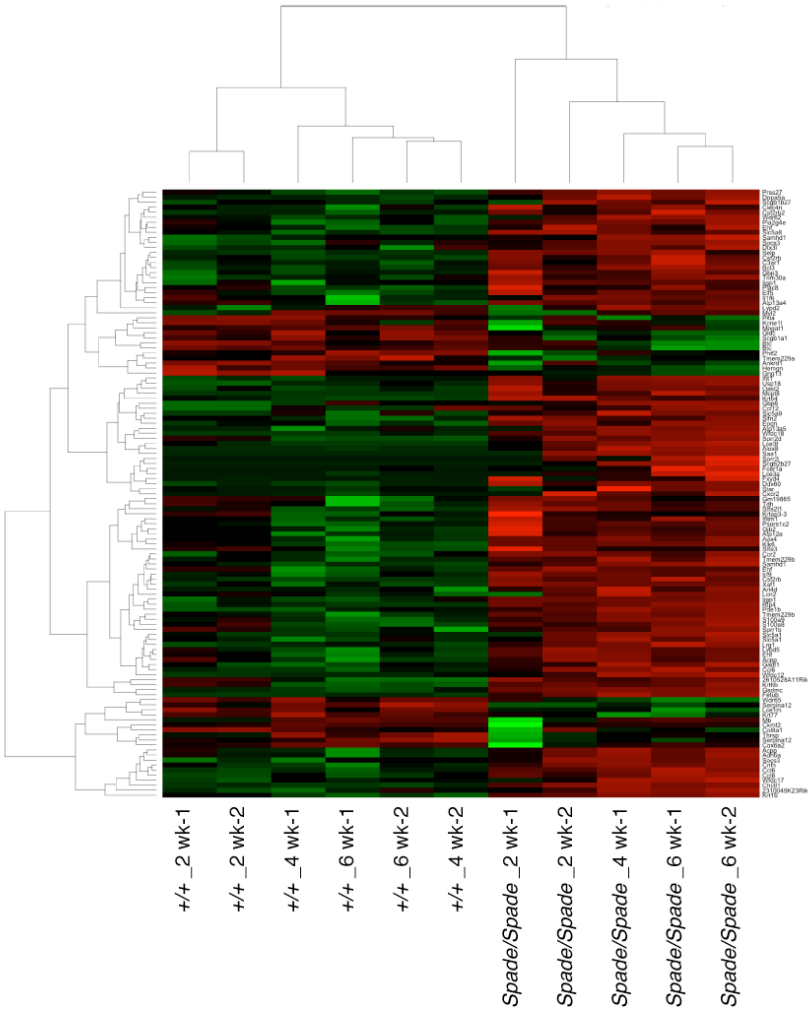
Supplemental Figure 8

Histological analysis of petrolatum-treated ear skin. Photomicrographs showing hematoxylin and eosin (HE) staining of ear skin histological sections from *Spade/Spade* and WT mice treated with a topical application of petrolatum (3 times a week from 4 weeks to 8 weeks of age) or left non-treated as a control. Scale bar, 100 μm; inset, 20 μm.



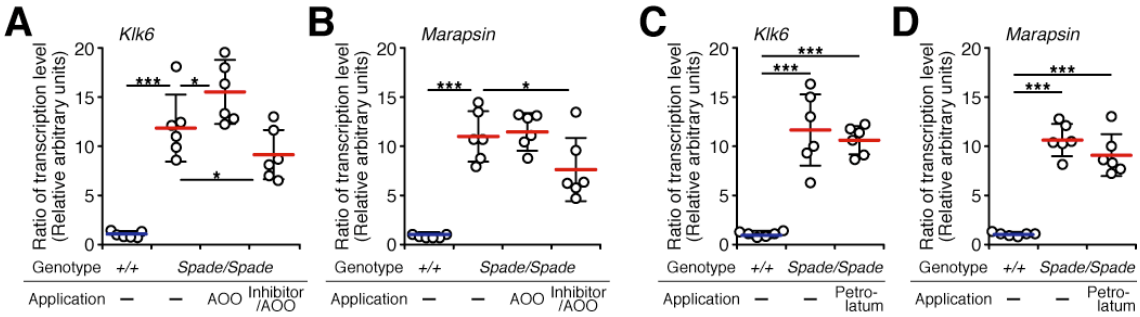
Supplemental Figure 9

Histological analysis of tape-stripped ear skin. Photomicrographs showing hematoxylin and eosin (HE) staining of ear skin histological sections from +/*Spade* and WT mice treated with a repeated application of cellophane tape (once a week from 8 weeks of age for 3 weeks) or left non-treated as a control. Scale bar, 100 μm.



Supplemental Figure 10

Heatmap of mRNA expressed in *Spade/Spade* and WT mice ear skin. Expression of whole skin tissue mRNA harvested from *Spade/Spade* and WT littermate was compared by mRNA microarray analysis and the genes upregulated or downregulated more than two fold in mutant skin are shown in this heatmap.



Supplemental Figure 11

Expression of mRNA after topical application of JAK inhibitor or petrolatum. Topical application of the JAK inhibitor I (**A** and **B**) or petrolatum (**C** and **D**) was performed 3 times a week for 3 weeks from 4 weeks of age. Ear tissues were sampled and mRNA expression levels of *Klk6* (**A** and **C**) and *Marapsin* (**B** and **D**) were examined. The results are expressed as fold transcript level relative to each untreated WT mouse. Each circle represents the ratio of the transcript level of individual mice, and bar shows the mean ± SD. **P* < 0.05, ****P* < 0.001, 2-tailed Student's t-test.