Supplemental information

[Title]

Signs of atopic dermatitis and contact dermatitis affected by distinct *H2*-haplotype in the NC/Nga genetic background

[Short Title]

AD sensitivity by distinct *H2*-haplotype in the NC/Nga background

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[Keyword] H-2^{nc}, *H2*-congenic NC/Nga mice, DNFB, atopic dermatitis (AD), allergic contact dermatitis (ACD)

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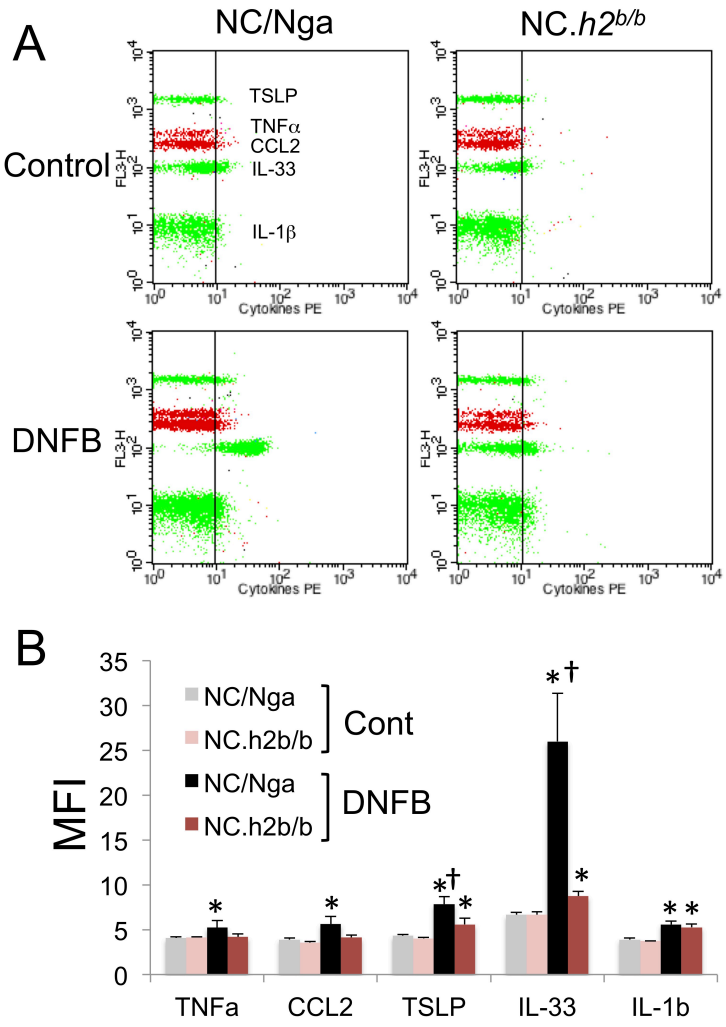
Supplementary Figure



Supplemental Figure S1. The expression pattern of serum cytokines in NC/Nga and NC.*h2*$^{b/b}$ were analyzed by flow cytometry. **A.** A typical DATA of flow cytometry is shown. **B.** The bar graphs (n = 4) depicting the mean fluorescence intensity (MFI) are indicated as mean ± SD. * DNFB *vs.* control, $p < 0.05$, *ANOVA*. † NC/Nga *vs.* NC.*h2*$^{b/b}$, $p < 0.05$, *ANOVA*

Supplemental Material

Cytokines concentration in serum was determined using LEGENDplex™ Multi-Analyte Flow Assay Kit (BioLegend, TNFα-A7, CCL2-A8, IL-1β-B2, IL-33-B5, and TSLP-B9) according to the manufacturer's instructions.[1] Briefly, two size populations of bead A and bead B for the microsphere populations are distincted by FSC/SSC analysis. Each beads have a characteristic fluorescence intensity that can be detected with FL3, and were coated by each cytokine-specific mAbs. Serum cytokines are sandwiched with biotinylated each cytokine-specific mAbs on each mixed beads, followed to labeling with streptavidin-PE. The intensities in the classification fluorescence channels which are easy to gate and quantify are clearly defined by FL3/FL2 assays.

Reference

1. Lehmann J.S., *et al.* Multiplex cytokine profiling of stimulated mouse splenocytes using a cytometric bead-based immunoassay platform. *J Vis Exp.* 129: e56440. doi: 10.3791/56440 (2017)