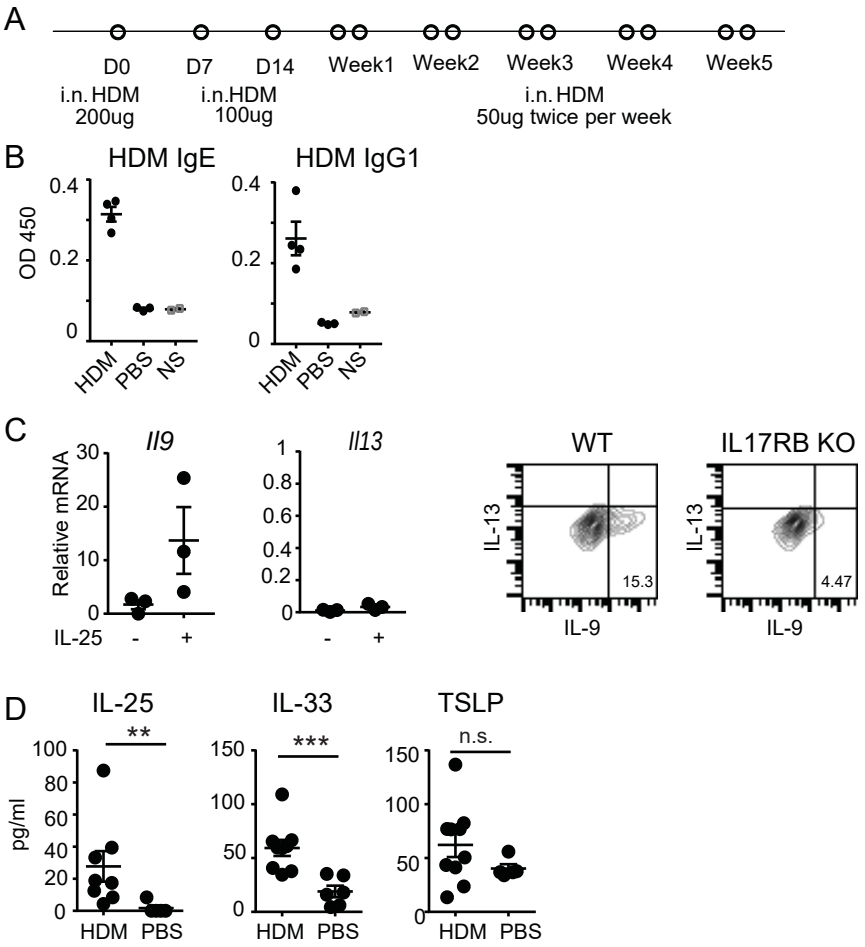
Suppl. Figure 1



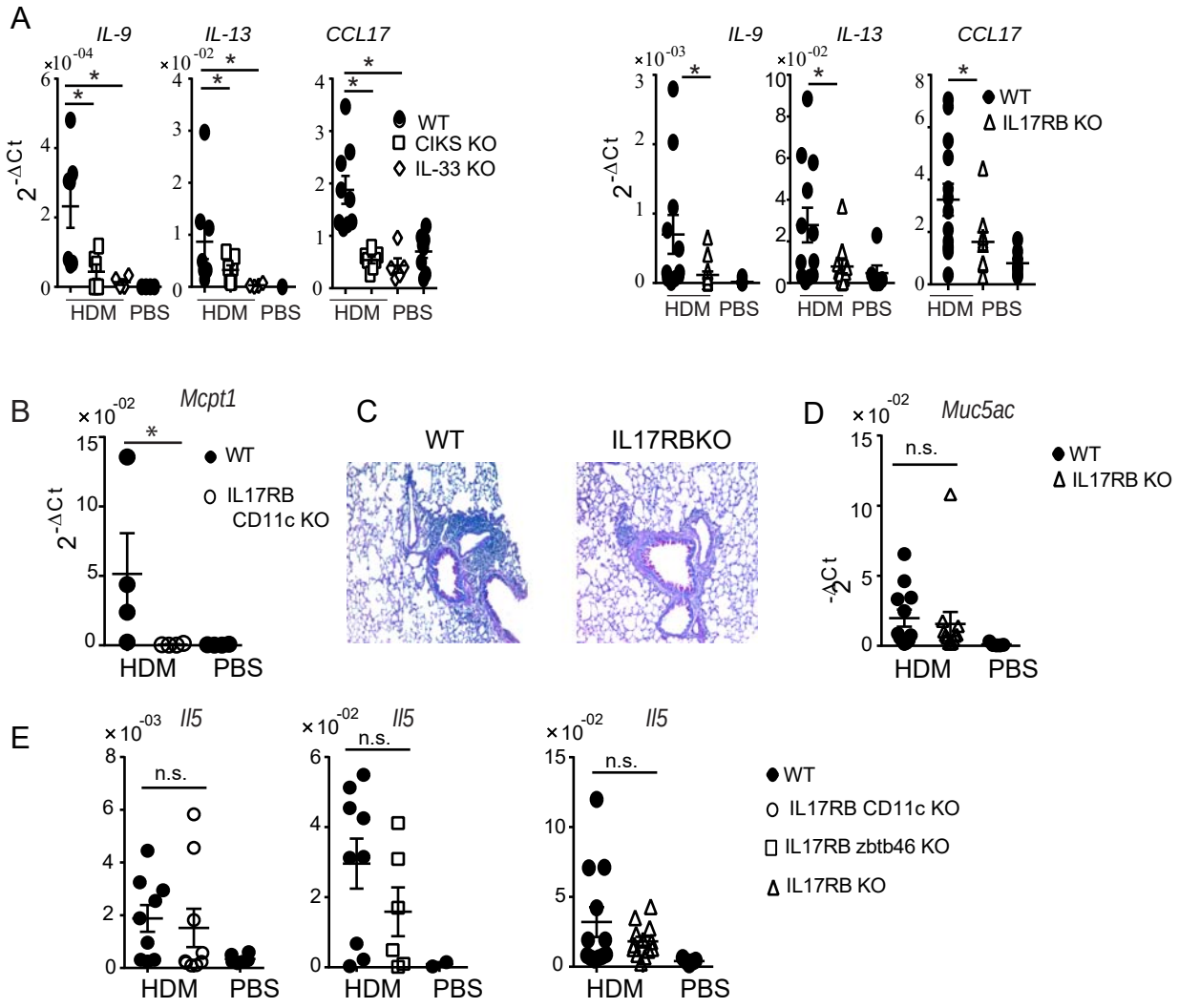
(A) Protocol for chronic HDM inhalation model of asthma.

(B) HDM induces anti-HDM immunoglobulins. Serum anti-HDM IgE and IgG1 24h after last inhalation in HDM or PBS control treated mice, (NS no serum). Data are the mean ± sem of n=3-4 mice from one experiment. For ELISA, plate was coated with HDM and developed with SBA Clonotyping System-HRP (SouthernBiotech, Birmingham AL).

(C) IL-25 induces IL-9 production of Th9 T cells. (Left panels) *Il9* and *Il13* mRNA levels after Th9 differentiation with or without IL-25 (100 ng/ml, Biolegend, San Diego, CA). Data are the mean ± sem of three independent experiments. (Right panels) Representative FACS data of three independent experiments.

(D) IL-25, IL-33 and TSLP levels at end of HDM treatments. Protein levels of IL-25, IL-33 and TSLP in lungs at the end of the HDM treatment regimen. Data are the mean ± sem of pooled data two from independent experiments (n=6-9 mice). IL-25 ELISA from Biolegend (Dedham MA), IL-33 and TSLP ELISAs from Thermo Fisher (Waltham MA)

Suppl. Figure 2



(A) IL-25 signaling pathway regulates *Il9*, *Il13* and *Ccl17* mRNA expression in lung in chronic HDM model. *Il9, Il13* and *Ccl17* relative expression in lungs from HDM or PBS treated WT and mutant mice, as indicated. Data are the means ± sem of pooled data from two independent experiments with total n=6-9 mice per group (left panel) and from three independent experiments with total of n=7-12 mice per group (right panel)

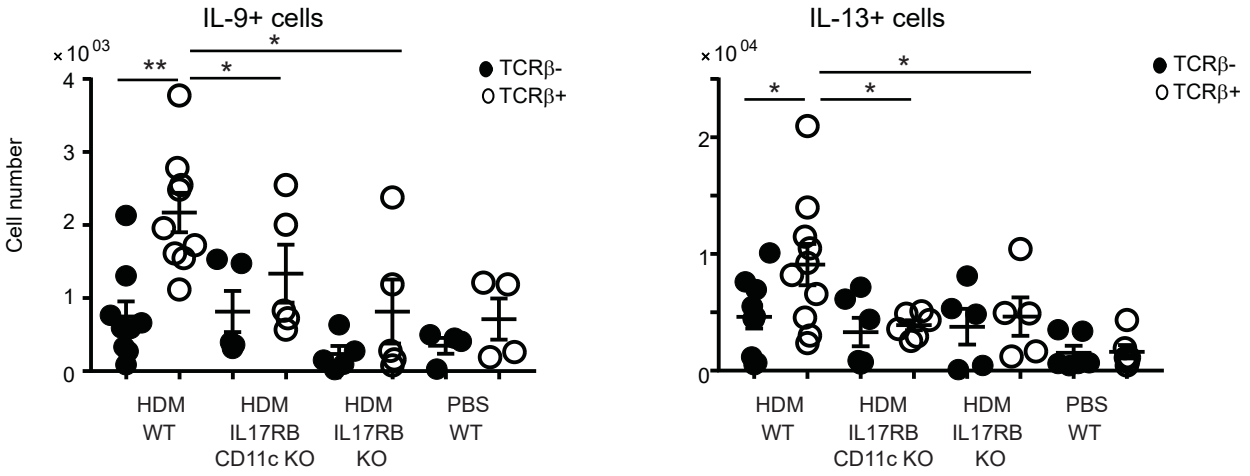
(B) HDM-induced mast cell accumulation. *Mcpt1* RNA levels in lungs of IL-17RB CD11c KO and WT mice after HDM or PBS exposure. Data are the mean ± sem of one experiment with total of n=4 mice per group

(C, D) HDM-induced mucus production is largely independent of IL-25. () Representative PAS stained lung sections of HDM-treated WT and IL17RBKO mice. For mucus analysis, sections were stained with periodic acid Schiff (PAS) (Millipore Sigma, St. Louis, MO). Muc5a mRNA levels in HDM or PBS treated lungs from WT or IL17RB KO. Data are the mean ± sem of three independent experiments with total of n=5-10 mice per group

(E) IL-25 signaling in lung CD11c does not regulated *Il5* expression in lung after chronic exposure to HDM. Relative *Il5* mRNA in lungs of IL17RB CD11c KO, IL17RB zbtb46 KO and IL17RB KO compared to WT mice. Data are the mean ± sem of two independent experiments with total of n=6-10 mice per group for IL17RB CD11c KO and IL17RB zbtb46 KO and of three independent experiment with total of n=6-12 for IL17RB KO.

*p<0.05; n.s. not significant

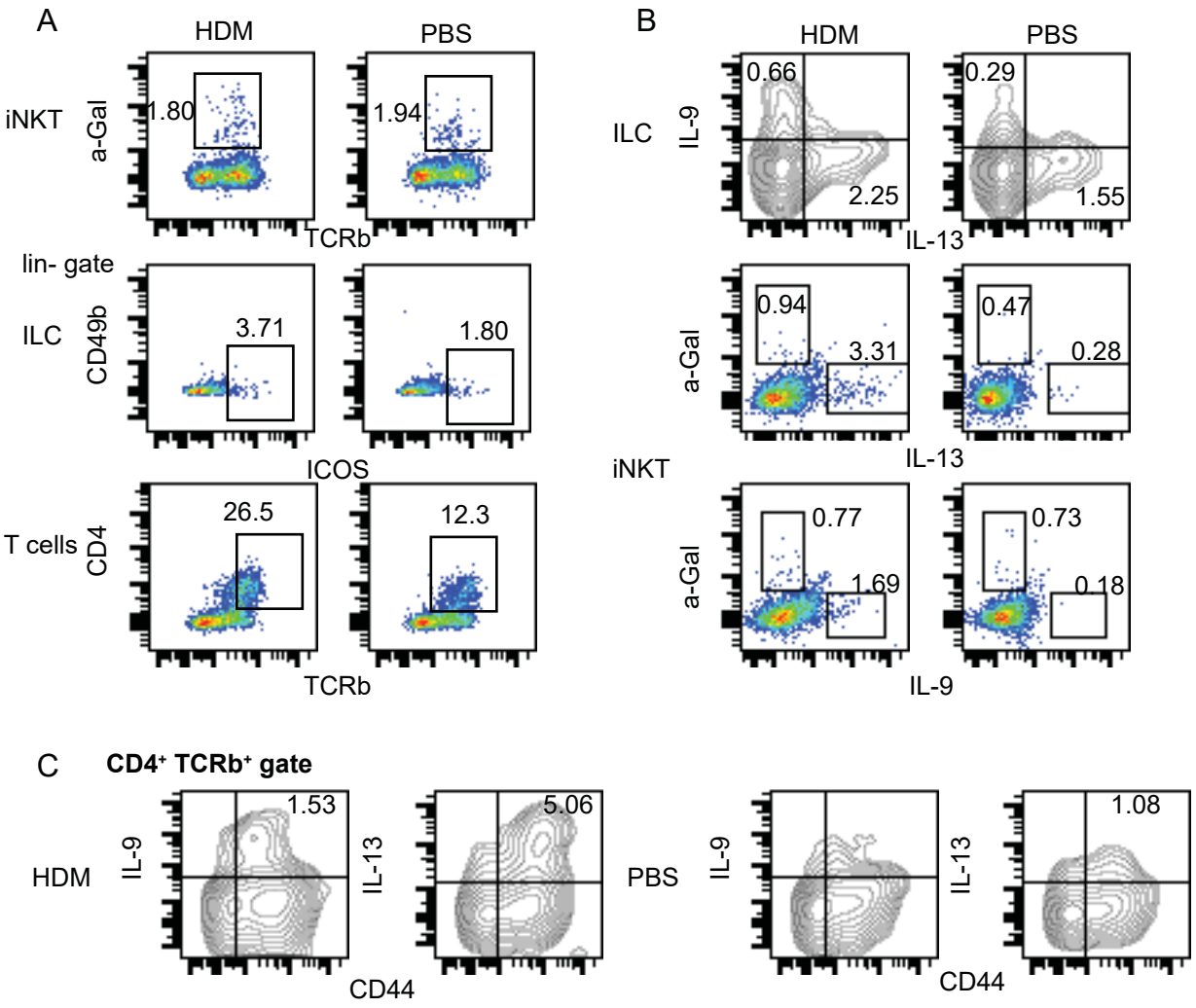
Suppl. Figure 3



TCRβ⁺cells are primary sources of IL-9 and IL-13 in chronic HDM model, partly dependent on IL-25-signaling pathway in CD11c⁺ cells

Numbers of IL-9 (left panel) and IL-13 producing (right panel) TCRβ⁺ and TCRβ⁻ cells in lungs from HDM or PBS treated WT and mutant mice, as indicated. Data are the means ± sem of pooled data from two independent experiments with total of n=10 for WT, n=6 for IL17RB CD11c KO, and n=5 for IL17RB KO n=5. *p<0.05; **p<0.01

Suppl. Figure 4



Effects of chronic HDM exposures on cellular composition and cytokine expression in lungs

For FACS analysis, cells were first gated on FSCvs SSC to eliminate debris, then on single cells (FSC-A vs FSC-H) and lastly, dead cells were excluded with aqua staining.

(A) Representative FACS analyses indicating percentage of iNKT, ILC2 and αβT cells in lungs of WT mice after chronic HDM or PBS exposure. iNKTs (TCRβ$^+$, αGalactosylceramide tetramer$^+$), ILCs (Lin$^-$ CD49d$^-$ ICOS$^+$) and CD4 T cells (TCRβ$^+$ CD4$^+$). (B,C) Representative FACS analyses showing IL-9 and IL-13 producing ILC2 and iNKT cells (B) and CD44$^+$ CD4$^+$ αβT cells (C) in lung with WT mice after chronic HDM or PBS exposure. Cells were stained with a combination of antibodies: APC-cy7 CD11c (HL3), APC-Cy7 CD4(GK1.5), APC-cy7 CD19 (1D3), PB CD11b (mac-1), PB- TCRβ (H57-597), Pe-cy7 CD49d (DX5), FITC-CD278 (ICOS), PercP CD44(BJ18), PB-CD8 (53-9.7) and APC-αGalactosylceramide-tetramer (NIH tetramer facility). All antibodies were purchased from BD Biosciencs (San Jose CA), Ebioscience (ThermoFisher, Waltham MA) and Biolegend (Dedham MA). For intracellular staining, cells were cultured with cell stimulation cocktail (Thermo fisher, waltham, MA) for 4h.

Data representative of analyses from two independent experiments with total of n=6 mice