SUPPLEMENTAL DATA



Supplemental Figure 1. Presence of T cells suppresses generation of CCR10⁺ NK1.1⁺ ILC1s in adult thymi. A, B, C) Flow cytometric (FC) analysis of CD3⁻NK1.1⁺ cells in thymocytes of adult Rag1^{-/-}CCR10^{+/EGFP} (A), TCR $\beta^{-/-}$ CCR10^{+/EGFP} (B) and TCR $\delta^{-/-}$ CCR10^{+/EGFP} (C) mice. Average percentages and number of CD3⁻NK1.1⁺ thymocytes were showed in bar graphs in comparison to their WT controls. CD3⁻NK1.1⁺ cells are gated from CD4⁻CD8⁻ thymocytes. Results are representative of two or three independent experiments. ns = not significant, **p < 0.01, ***p < 0.001, ****p < 0.0001 as determined by two-tailed Student's t test.



Supplemental Figure 2. Impaired $\alpha\beta T$ and $\gamma\delta T$ cell differentiation in DL4-KO thymi. A) FC analysis of thymocytes for expression of CD4 and CD8 in adult DL4-WT (DL4^{f/f} CCR10^{+/EGFP}) and DL4-KO (FoxN1^{Cre}DL4^{f/f}CCR10^{+/EGFP}) mice. **B**) Comparison of the percentage and number of thymic $\gamma\delta T$ cells in adult DL4-WT and DL4-KO mice. C) FC analysis of thymocytes for expression of CD4 and CD8 in newborn adult DL4-WT and DL4-KO mice. D) Comparison of the percentage and number of thymic $\gamma\delta T$ cells in newborn DL4-WT and DL4-KO mice. Results are of three independent experiments. ns = not significant and *p < 0.05, ***p < 0.001, ****p < 0.0001 as determined by two-tailed Student's t test.



Supplemental Figure 3. Differentiation of CCR10⁻ CD3⁻ NK1.1⁺ thymocytes into CCR10⁺ CD3⁻ NK1.1⁺ cells in the OP9-DL1 culture. FC analysis of the expression of CCR10(EGFP) on gated CD45⁺CD3⁻NK1.1⁺ cells generated from the *in vitro* co-culture of purified CCR10(EGFP)⁻ CD3⁻CD4⁻CD8⁻ NK1.1⁺ thymocytes with OP9 (dotted line) or OP9-DL1 (solid line) cells for 19 days. The CCR10(EGFP) histogram of CCR10^{+/+} cells (gray area) serves as a negative control for EGFP. Representative of three experiments.



Supplemental Figure 4. Analysis of CD103 and PLZF expression on CCR10⁻ and CCR10⁺ thymic NK1.1⁺ ILC1s of DL4-KO mice. A) Comparison of the PLZF expression on CCR10⁻ and CCR10⁺ CD3⁻NK1.1⁺ thymocytes in adult and newborn DL4-WT (DL4^{f/f} CCR10^{+/EGFP}), DL4-KO (Foxn1^{Cre}DL4^{f/f} CCR10^{+/EGFP}) and Rag1^{-/-}CCR10^{+/EGFP} mice. B) FC analysis of expression of EOMES, T-bet and CD11b in thymic NK1.1⁺ ILC1s of newborn DL4-WT and DL4-KO mice.