



Figure S1. Gating strategies. All are log10 scale. (A) Representative FACS analysis of progenitor T cells in control (Ctrl; top) and *Mx1-Cre p107-Single* (Mut; bottom) mice. Lineage-negative (CD45⁻, Mac1⁻, Ter119⁻, Sca1⁻) cells were stained for Thy1, cKit, and CD25. (B) Representative FACS analysis of mature T cells in control (top) and *Mx1-Cre p107-Single* (bottom) mice. Live cells were stained for either CD3 ϵ or CD4/CD8 markers. (C) Representative FACS analysis of naive splenic T cells in control (top) and mutant (bottom) mice. Splenocytes were first stained for CD4 and CD8, and both subpopulations of T cells were stained for CD62L and CD44. Naive T cells are CD62L⁺ CD44⁻ ($n = 5$). (D) Representative flow analysis of CD40⁻ and CD205⁺-labeled cTEC subsets in control (top) and mutant (bottom) thymi. (E) Representative flow analysis of UEA1⁻ and CD80⁺-labeled mTEC subsets in control (top) and mutant (bottom) thymi. FSC, forward scatter.