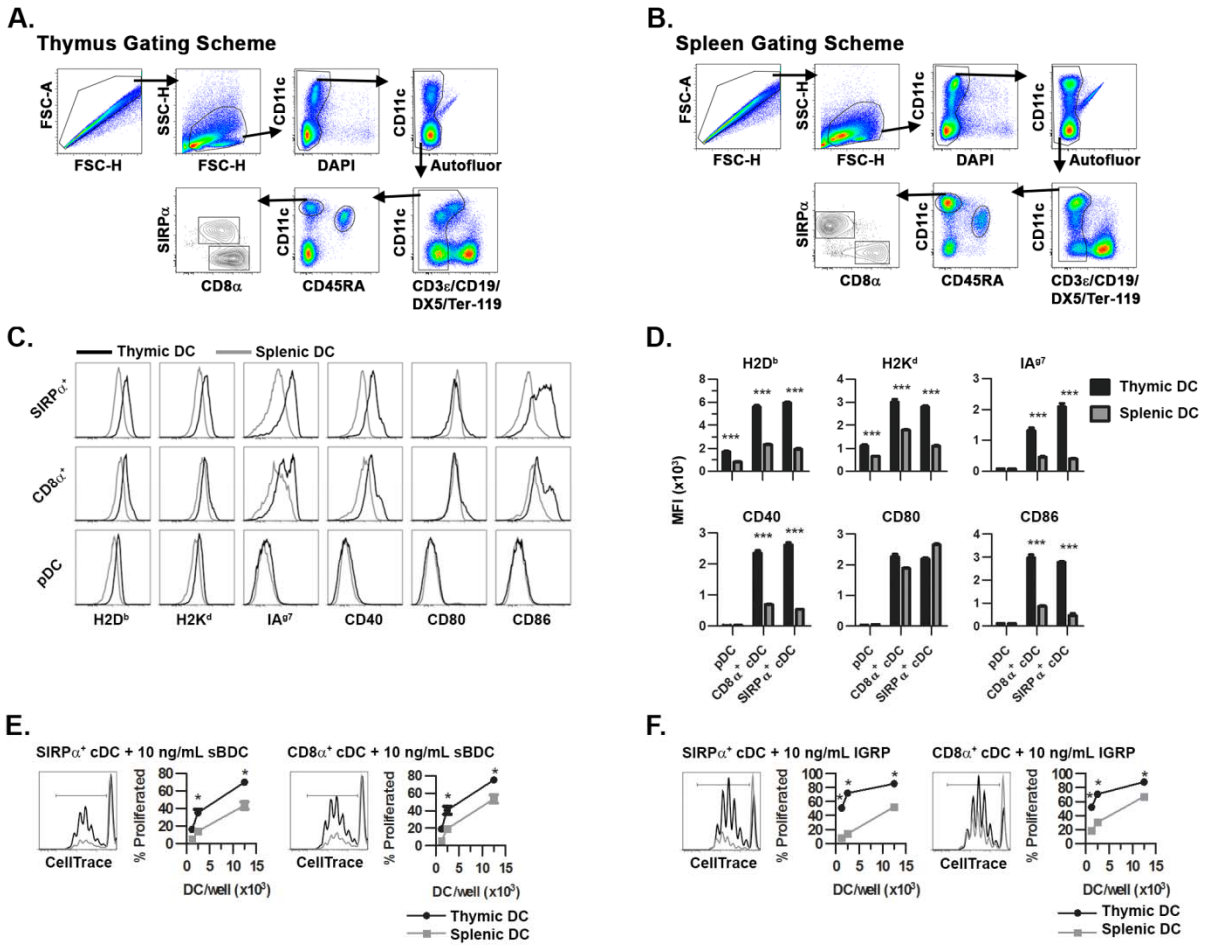
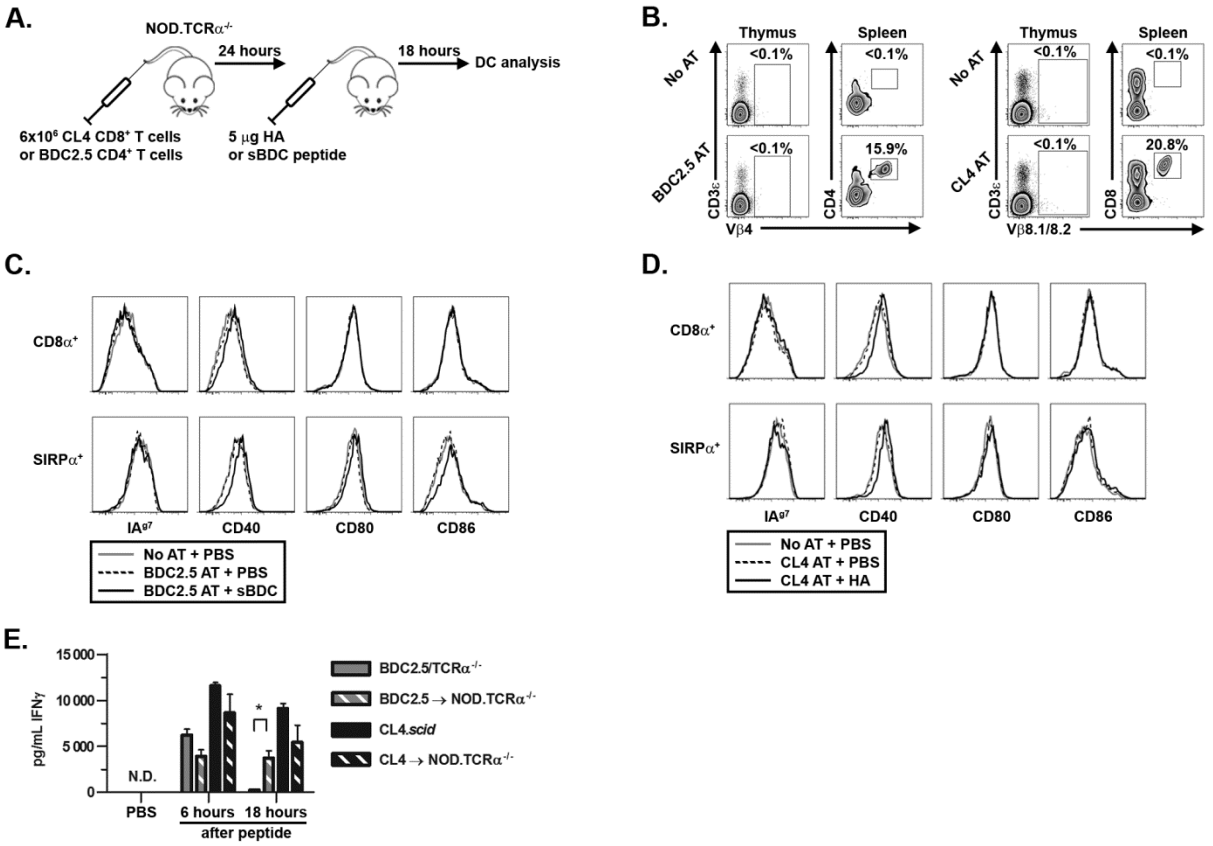


Supplemental Figure 1. NOD thymic DC are phenotypically and functionally more mature than splenic DC. Gating scheme for all analyses of DC from NOD mouse (A) thymus and (B) spleen following isolation as described in *Materials and Methods*. (C-D) Comparison of MHC I ($H2D^b$, $H2K^d$), MHC II (IA^{g7}), CD40, CD80, and CD86 expression by NOD thymic and splenic DC (n=4). Data are representative of at least 4 experiments. (E-F) Thymic and splenic cDC subsets were FACS-sorted, pulsed with (E) sBDC or (F) IGRP peptide, then co-cultured with (E) BDC2.5 $CD4^+$ or (F) 8.3 $CD8^+$ T cells for 3 d. Proliferation of live $Thy1.2^+CD4^+$ or $CD8^+$ T cells was determined via CellTrace Violet dilution. Plasmacytoid DC induced no proliferation under the conditions tested (data not shown). Graphed data are pooled from 2 independent experiments. *, $P<0.05$; ***, $P<0.001$ by two-way ANOVA with Bonferroni posttest.



Supplemental Figure 2. Activation of peripheral T cells alone does not activate thymic DC. (A) Experimental design to determine if peripheral T cell activation leads to thymic DC activation. (B) Detection of transferred T cells by characteristic TCR Vβ staining. Thymus panels were gated on total, live thymocytes; spleen panels were gated on live, CD3ε^{+}Thy1.2^{hi} cells. AT, Adoptive Transfer. (C-D) Analysis of thymic DC phenotype in NOD.TCRα$^{-/-}$ mice that received (C) BDC2.5 CD4^{+} T cells and 5 μg sBDC or (D) CL4 CD8^{+} T cells and 5 μg HA as described in panel A. (E) IFNγ concentration in the serum of T cell-injected NOD.TCRα$^{-/-}$ recipients or BDC2.5/TCRα$^{-/-}$ and CL4.*scid* mice injected with peptide (n=3-6/group). *, P<0.05 by two-way repeated measures ANOVA with Bonferroni posttest.