Supplementary Figure 1. Identification of M2a, M2b and M2c Subsets Within Mature cSP4 and SP8 Thymocytes

(A) Flow cytometric analysis of the frequency of TCR β^{hi} SP8 in+/*plt* (n=16) controls and *plt/plt* mice (n=16). (B) Flow cytometric analysis showing gating strategy for the detection of M2a, M2b and M2c subsets of mature CD69⁻CD62L⁺ cSP4 (A) and SP8 (B) in +/*plt* (n=16) controls and *plt/plt* mice (n=16). Flow cytometry data representative of at least 3 independent experiments. In all cases, error bars represent mean +/- SEM; ****p<0.0001



Supplementary Figure 2. CCL19 Is Dispensable For Neonatal SP Thymocyte Egress

Panel (A) show flow cytometry and quantitation of CD4/CD8 thymocyte subsets in P10 *Ccl19*^{-/-} (white bars) and *Ccl19*^{+/-} (black bars) littermate controls. Quantitation of immature CD69⁺CD62L⁻ and mature CD69⁻CD62L⁺ cSP4 (panel B) and TCR β^{hi} SP8 (panel C) thymocytes, in *Ccl19*^{-/-} (n=7, black bars) and *Ccl19*^{+/-} (n=10, white bars) littermate P10 neonatal mice. (D) shows frequencies of M2a, M2b and M2c subsets of mature CD69⁻CD62L⁺ cSP4 and SP8 in *Ccl19*^{+/-} (n=7, black bars) controls and *Ccl19*^{-/-} mice (n=10, white bars). Error bars represent mean +/- SEM. For analysis of data in D, multiple comparison analysis was achieved by a Two-way ANOVA followed by Sidak's post-test in GraphPad Prism to determine statistical differences. Flow cytometry data representative of at least 2 independent experiments.



Supplementary Figure 3. CCL21-Deficient Thymic Grafts Contain Conventional SP Thymocyte Populations

Panel (A) shows flow cytometric analysis of CD4, CD8 expression in $Ccl21a^{+/-}$ or $Ccl21a^{-/-}$ grafted thymus lobes, harvested 7 days after surgery. Also shown are numbers of total thymocytes, CD4⁺CD8⁺ thymocytes, and cSP4 and SP8TCR β^{hi} thymocytes in $Ccl21a^{+/-}$ (black bars) or $Ccl21a^{-/-}$ (white bars). Error bars represent mean +/- SEM, with a minimum of n=3 for each strain.

