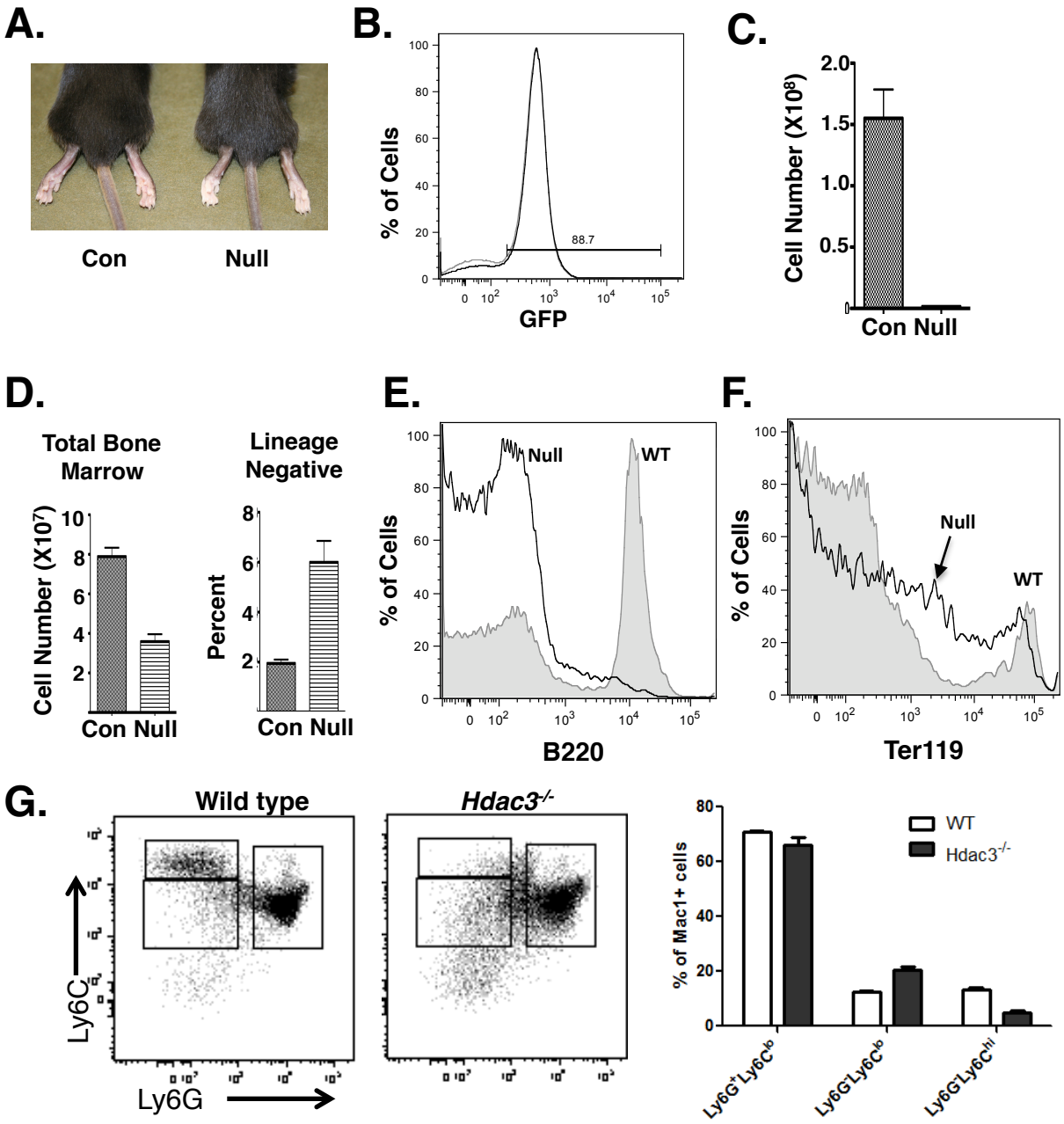
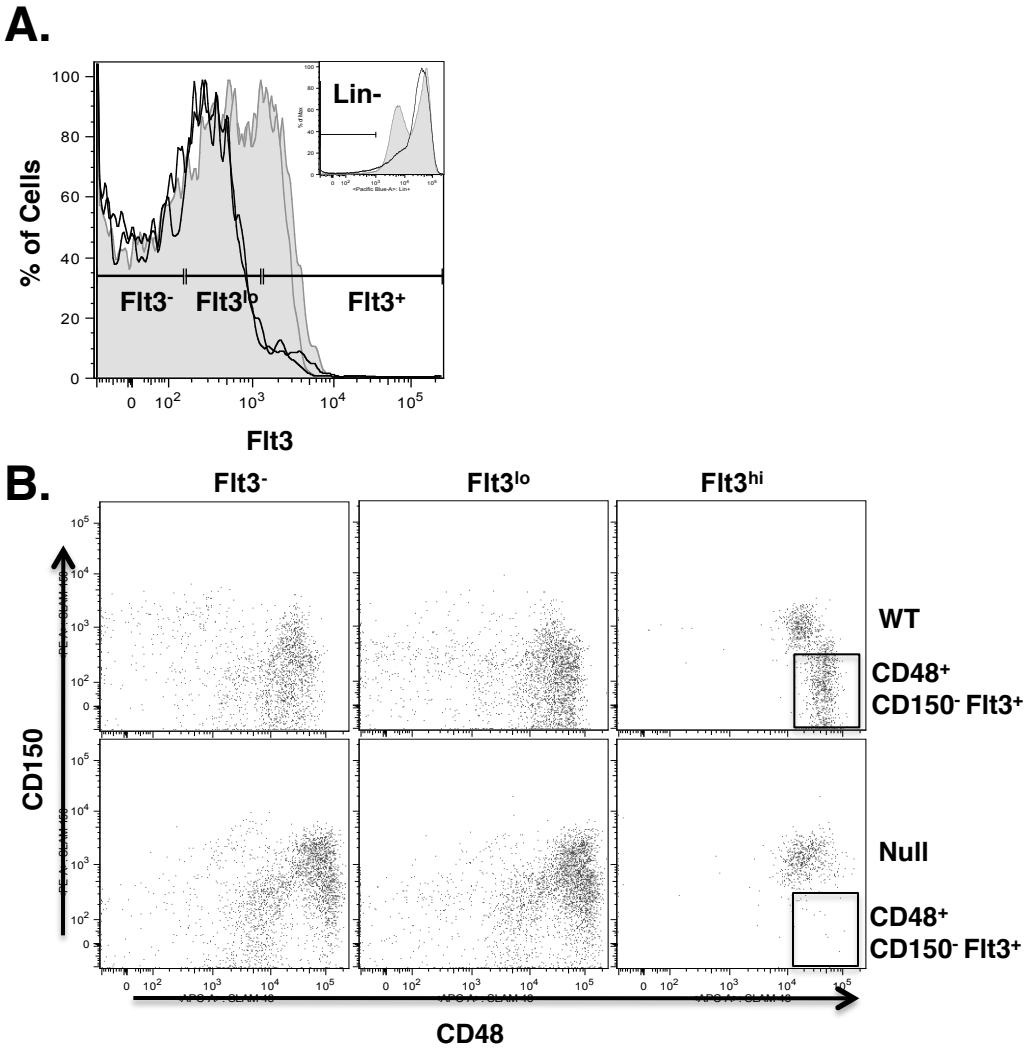
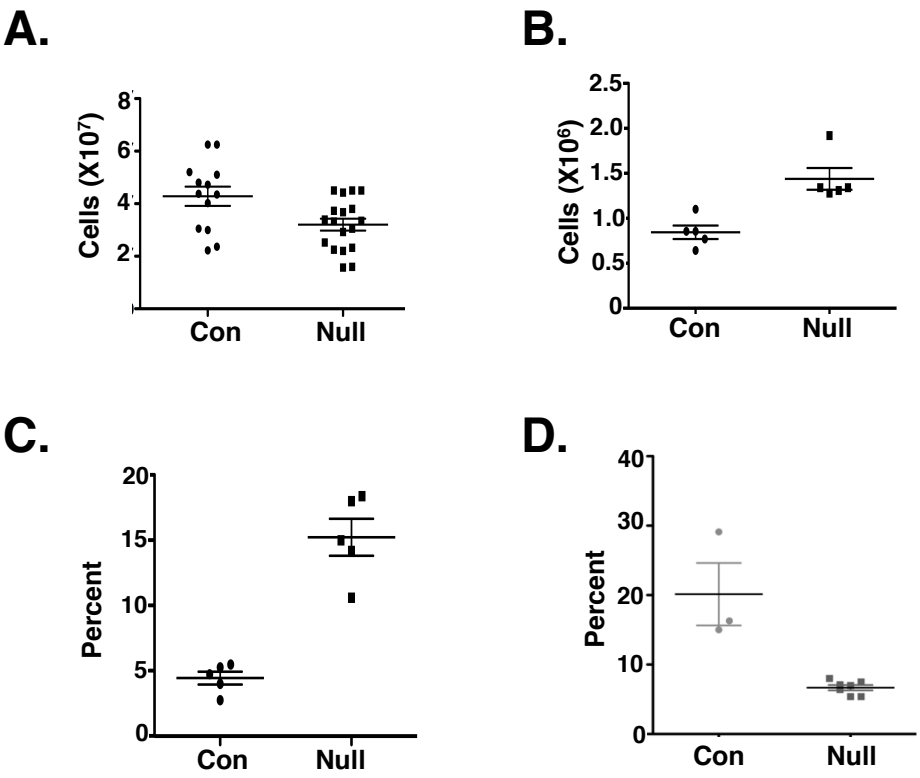


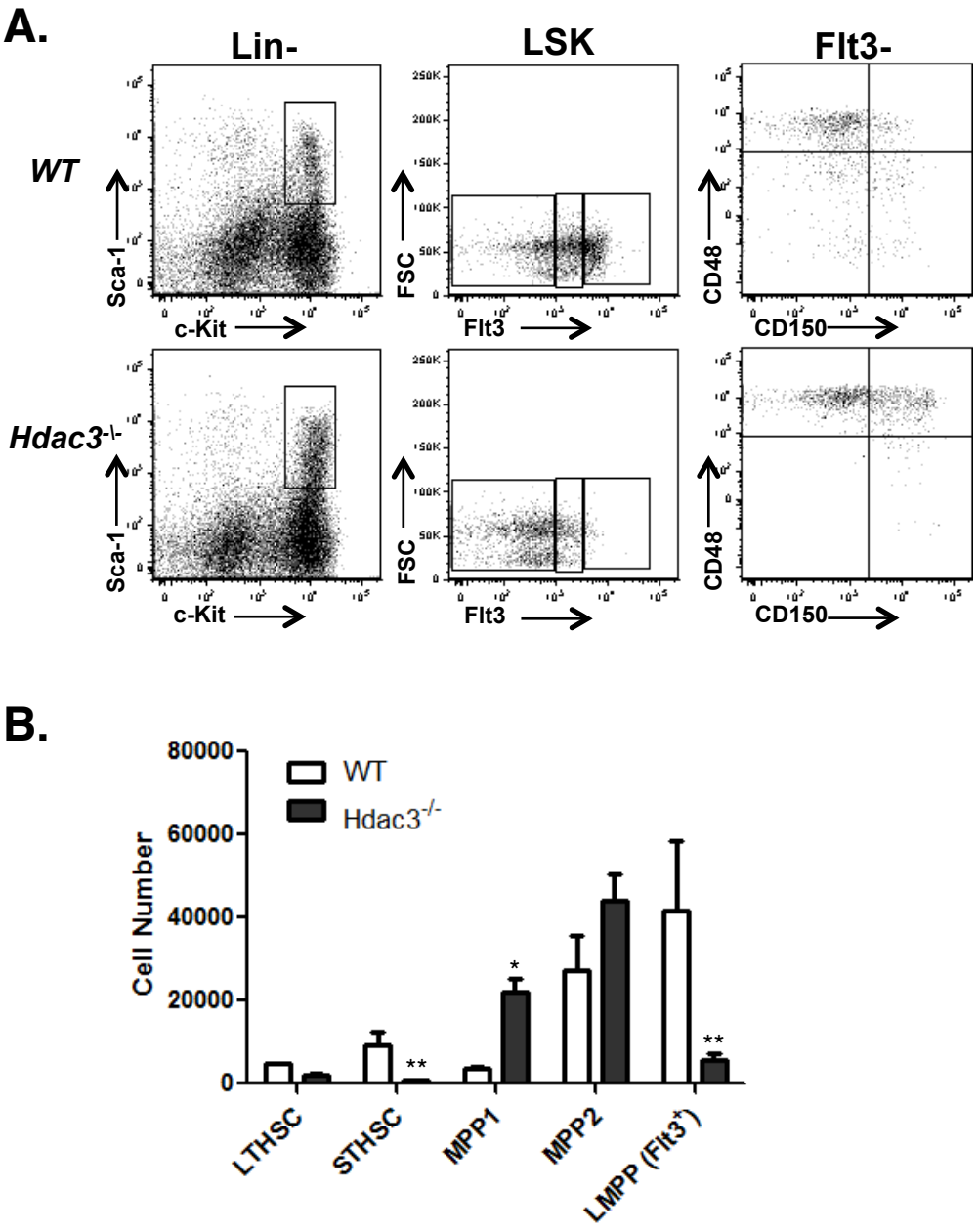
Supplemental Figure 1. Characterization of *Vav-Hdac3*$^{-/-}$ mice. (A) Photograph of control and *Vav-Hdac3*$^{-/-}$ mice showing the pale feet observed in the null mice. (B) Cre recombinase activity assessed by flow cytometry for GFP^{+} cells from the *ROSA26-lox-STOP-lox-GFP* allele. The number is the relative percentage of the cells in the indicated gated population. (C) Quantification of number of thymocytes from control versus *Vav-Hdac3*$^{-/-}$ mice (n=5; *P = >0.05*). (D) Quantification of total numbers of bone marrow cells in control verses *Vav-Hdac3*$^{-/-}$ bone marrow and percent of total bone marrow that were lineage negative identified by staining with anti-CD3, anti-B220, anti-Mac-1, anti-Gr-1 and anti-Ter119 (n=5; *P = >0.05*). (E & F) FACS analysis of total bone marrow assessed for B-cells using anti-B220 (E) and erythrocytes using anti-Ter119 (F). Shown are representative FACS plots from at least 5 mice. (G) Mac1^{+} cells from control or *Mx1-Cre:Hdac3*$^{-/-}$ bone marrow were further fractionated using anti-Ly6C and Ly6G. FACS plots at the right show the gates used for the graphs at the left (Ly6G^{hi}Ly6C^{lo}, *P* = 0.36; Ly6G^{-}Ly6C^{lo}, *P* = 0.015; Ly6G^{-}Ly6C^{hi}, *P* = 0.005).



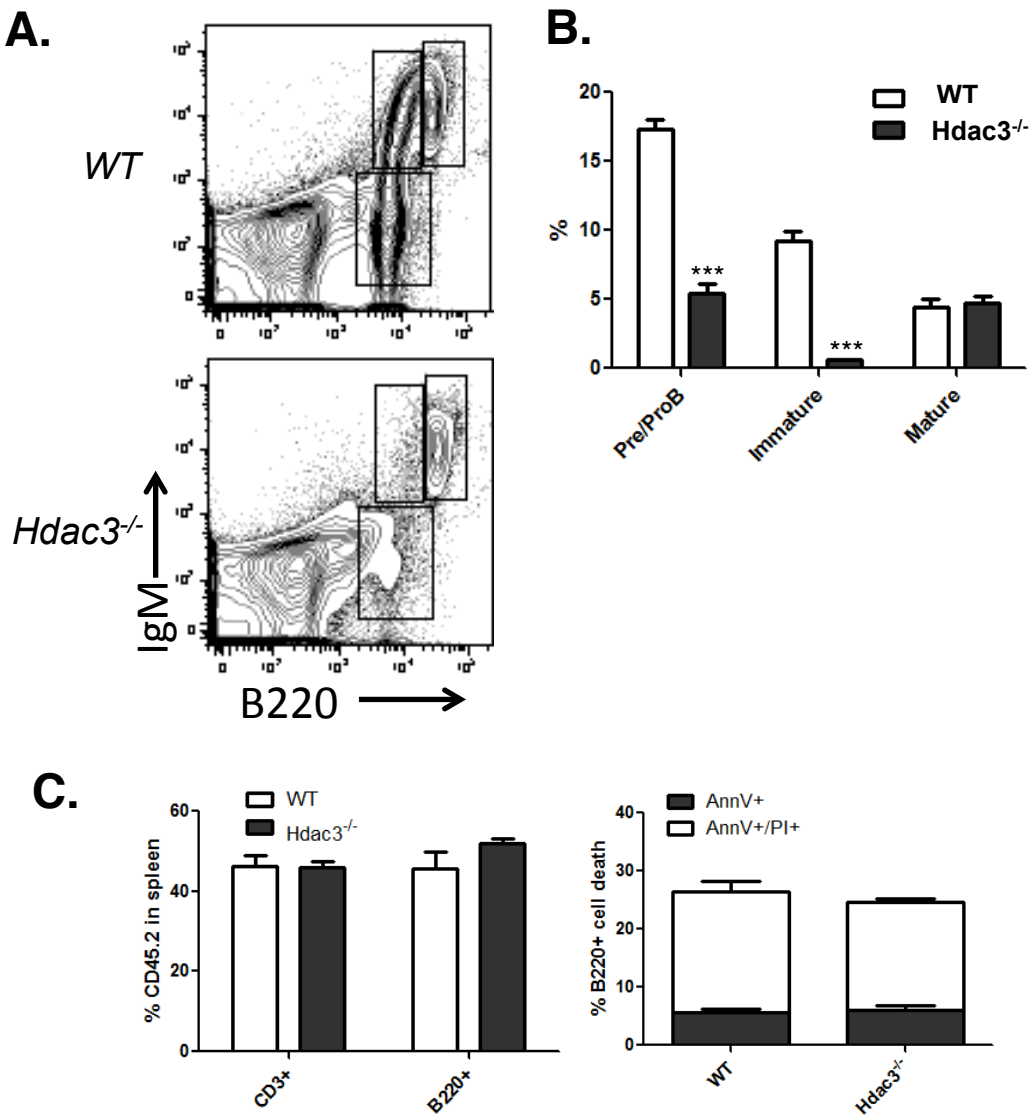
Supplemental Figure 2: Deletion of *Hdac3* results in block of progression of stem cells to LMPP. (A) FACS analysis of Flt3 expression from lineage negative populations from 2 control (shaded) and 2 null (black line) mice showing the specific Flt3 expression populations analyzed for CD150 and CD48 expression in (B). (B) FACS plots of two control and two null mice overlayed to show this small population of cells ($Flt3^+/CD150^-/CD48^+$). Shown is a representative experiment that has been done with 2 or more mice 3 times).



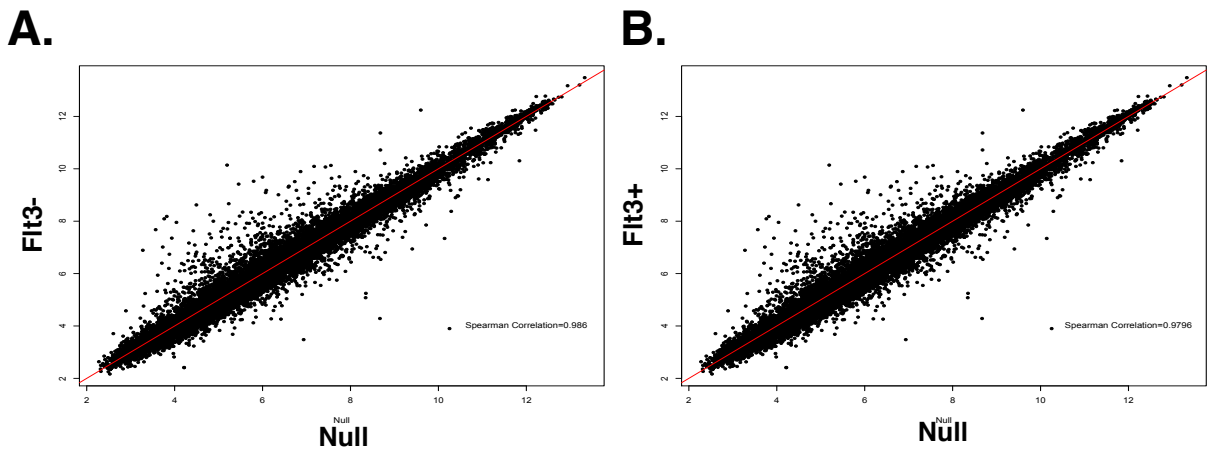
Supplemental Figure 3. Characterization of *Mx1-Hdac3*$^{-/-}$ mice. Quantification of total numbers of bone marrow cells in control verses *Mx1-Hdac3*$^{-/-}$ bone marrow **(**A; p = 0.0133) and cells that were lineage negative (B) identified by staining with anti-CD3, anti-B220, anti-Mac-1, anti-Gr-1 and anti-Ter119 (p = 0.0031). (C) FACS analysis of total bone marrow assessed for lineage negative, Sca-I^{+}, c-Kit^{+} cells that were also GFP+ (p = 0.01). (D) FACS analysis of B cells from the bone marrow of wild type and *Mx1-Hdac3*$^{-/-}$ mice using anti-B220 (p=0.0012).



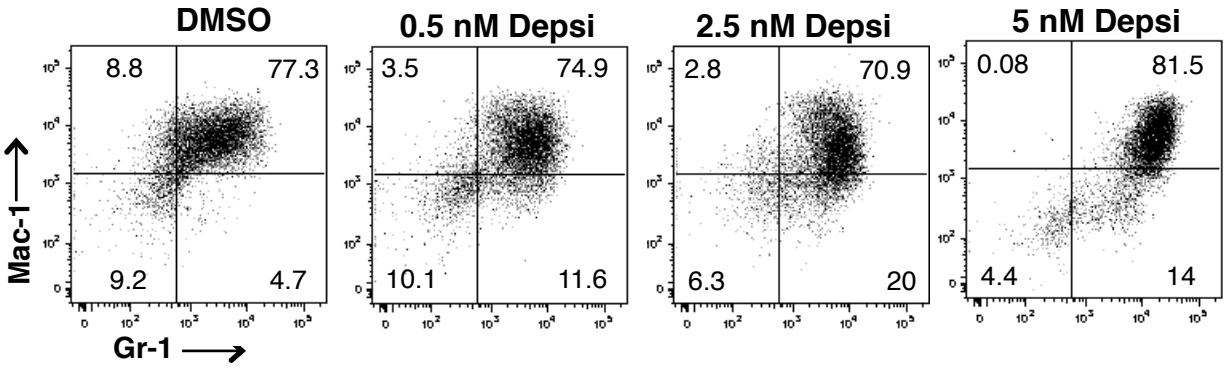
Supplemental Figure 4. *Hdac3*$^{-/-}$ hematopoietic defects are intrinsic to hematopoietic cells. Bone marrow from *Mx1-Hdac3*$^{-/-}$ mice was transplanted into wild type mice to create a tissue-specific deletion of Hdac3 only in hematopoietic cells. (A) Representative FACS analysis showing that the phenotype observed in Vav-Cre mice is also observed after poly I-C induction of *Mx1-Hdac3*$^{-/-}$ transplanted mice. (B) Quantification of total numbers of bone marrow cells in control verses *Mx1-Hdac3*$^{-/-}$ transplanted mice in which 2 control mice were compared to 6 *Mx1-Hdac3*$^{-/-}$ transplanted mice (STHSC, p=0.0018; MPP1, p=0.0228; LMPP, p=0.0052)



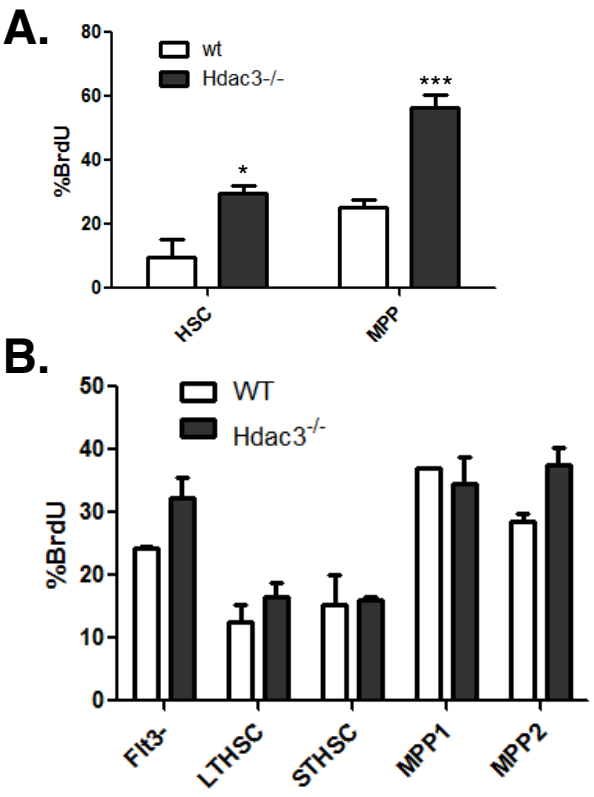
Supplemental Figure 5. Block of B cell development in *Mx1-Hdac3*$^{-/-}$ mice. FACS analysis of B220+ versus IgM+ B cells from the bone marrow of control and Hdac3-/- mice. (A) Representative FACS plots. (B) Quantification of biological replicates (WT, n=2; Hdac3$^{-/-}$, n=4) of FACS analysis of B cells from the bone marrow of wild type and *Mx1-Hdac3*$^{-/-}$(Pro/PreB, p=0.0003; Immature, p<0.0001). (C) Inducible *Hdac3* deletion does not affect splenic T or B cells (left panel) or induce apoptosis in the B220+ B cells in the bone marrow (right panel).



Supplemental Figure 6. Comparison of null samples and Flt3$^+$ sample at the whole transcriptome level. Scatterplot of gene expression between null samples and wild type Flt3$^-$ (A) or Flt3$^+$ (B)**.** Each point denotes one gene whose expression was expression was detectable. Red line is the 45 degree line. The Spearman correlation coefficient is 0.986 (A) and 0.980 (B), which is very high and indicates that the rankings of individual gene expression levels for null and Flt3$^+$ or Flt3$^-$ samples are consistent.



Supplemental Figure 7. Dose-dependent myeloid differentiation effects of Depsipeptide. FACS analysis of wild type LSK cells that were FACS sorted and cultured for 7 days on stromal OP9 cells in the presence of increasing concentrations of Depsipeptide. The loss of the double negative population occurred beginning at 2.5 nM, but stimulation of $Gr1^{+}$ cells began with as little as 0.5 nM. The numbers in each box are the relative percentage of the cells in the indicated gated population.



Supplemental Figure 8. BrdU incorporation in *Hdac3*$^{-/-}$ stem and progenitor cellls. (A) *Vav-Cre/Hdac3*$^{-/-}$ mice were injected with BrdU 2 hr prior to analysis for BrdU incorporation using FACS within the LSK/CD150^{+}/CD48^{-} (HSC) or LSK/CD48^{+} populations (MPPs) HSC, p=0.0144; MPP, p=0.0006. (B) A similar analysis to that in A but using *Mx1-Cre/Hdac3*$^{-\backslash-}$ mice lacking GFP expression to allow the inclusion of Flt3 to further segregate the populations as shown in Fig. 2E. While similar trends were found, these did not reach statistical significance; *WT*, n = 2; *Hdac3*$^{-/-}$, n = 5.