

SIRT1 is dispensable for maturation of hematopoietic stem cell in the bone marrow niche

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Figure S1. Semi-quantitative PCR was performed on genomic DNA from the cells of the indicated genotypes. Sirt1, sirtuin 1; Sirt1^{ΔΔ}, sirtuin 1 conditional knockout; Sirt1^{+/+}, sirtuin 1 wild-type. Cre, cre recombinase.

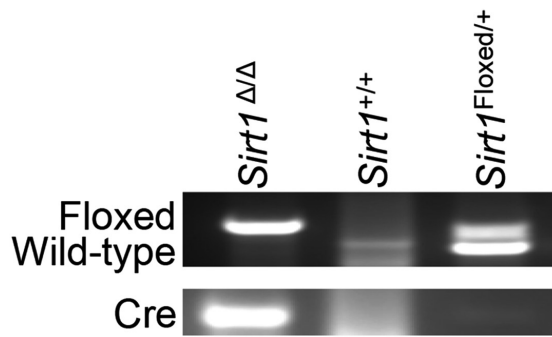


Figure S2. Representative flow cytometry plots of myeloid cells (Mac1⁺), B cells (B220⁺), and T cells (CD3⁺) from the BM from Figure 2 are shown. Mac1, macrophage-1 antigen.

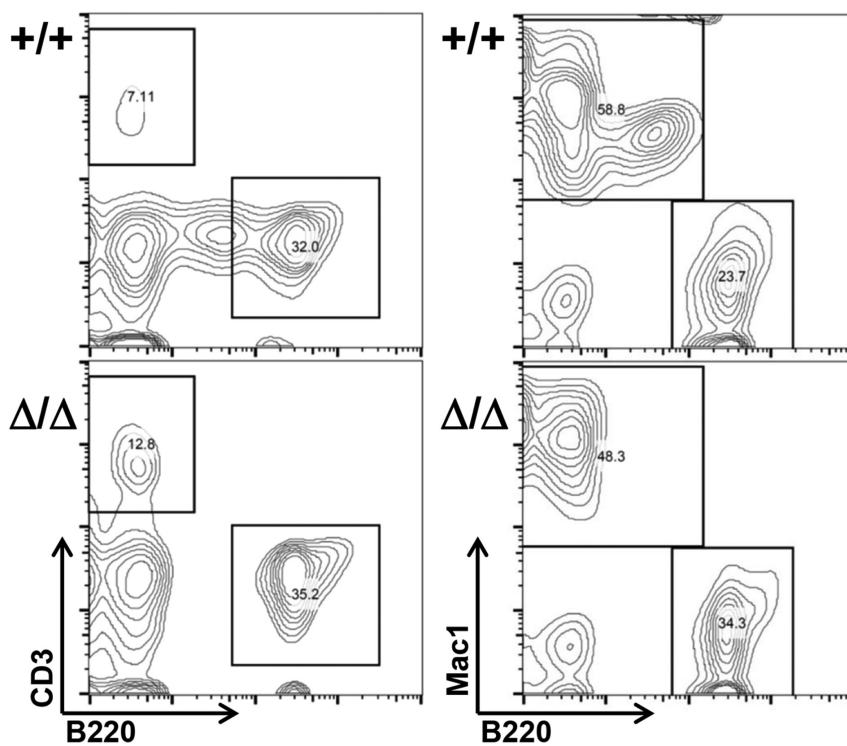


Figure S3. Representative flow cytometry plots of LSK cells, HSC, CLP, CMP, GMP and MEP from Figure 3 are shown. Lin, lineage; Sca1, spinocerebellar ataxia type 1; cKit, mast/stem cell growth factor receptor kit; HSC, hematopoietic (Lin⁻Sca1⁺cKit⁺CD150⁺CD48⁻) stem cells; LSK, Lin⁻Sca1⁺cKit⁺ cells; CMP, common myeloid progenitor (Lin⁻Sca1⁻cKit⁺CD34⁺CD16/32⁻) cells; GMP, granulocyte-macrophage progenitor (Lin⁻Sca1⁻cKit⁺CD34⁺CD16/32⁺) cells; MEP, megakaryocyte-erythroid progenitor (Lin⁻Sca1⁻cKit⁺CD34⁺CD16/32⁻) cells; CLP, common lymphoid progenitor (Lin⁻Sca1^{low}cKit^{low}CD127⁺) cells. BM, bone marrow.

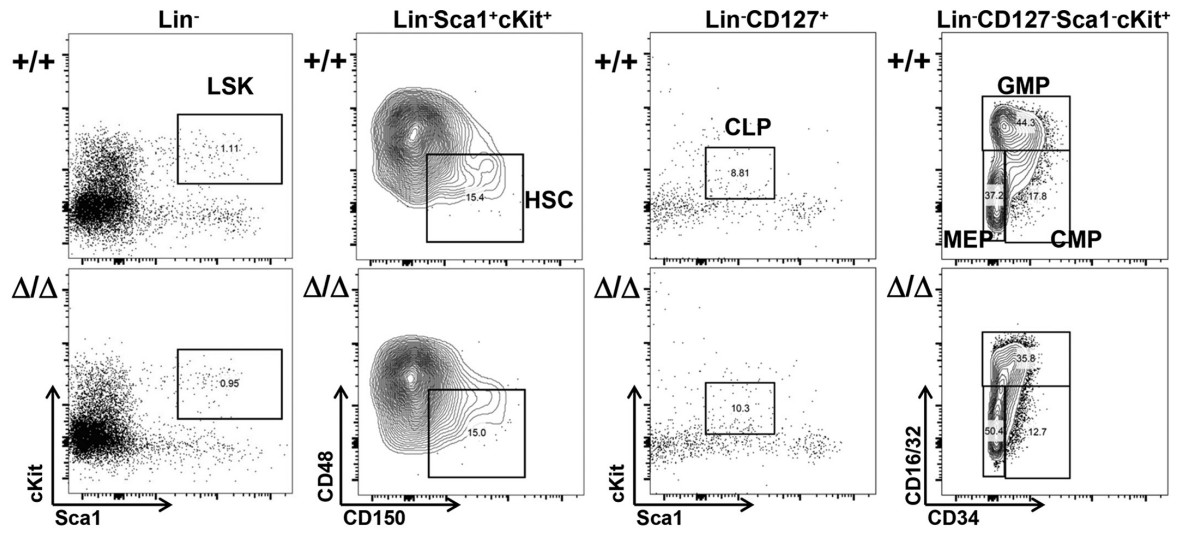


Figure S4. Representative flow cytometry plots of CD45.2⁺, myeloid cells (Mac1⁺), B cells (B220⁺), and T cells (CD3⁺) from the BM cells from Figure 4 are shown. Mac1, macrophage-1 antigen.

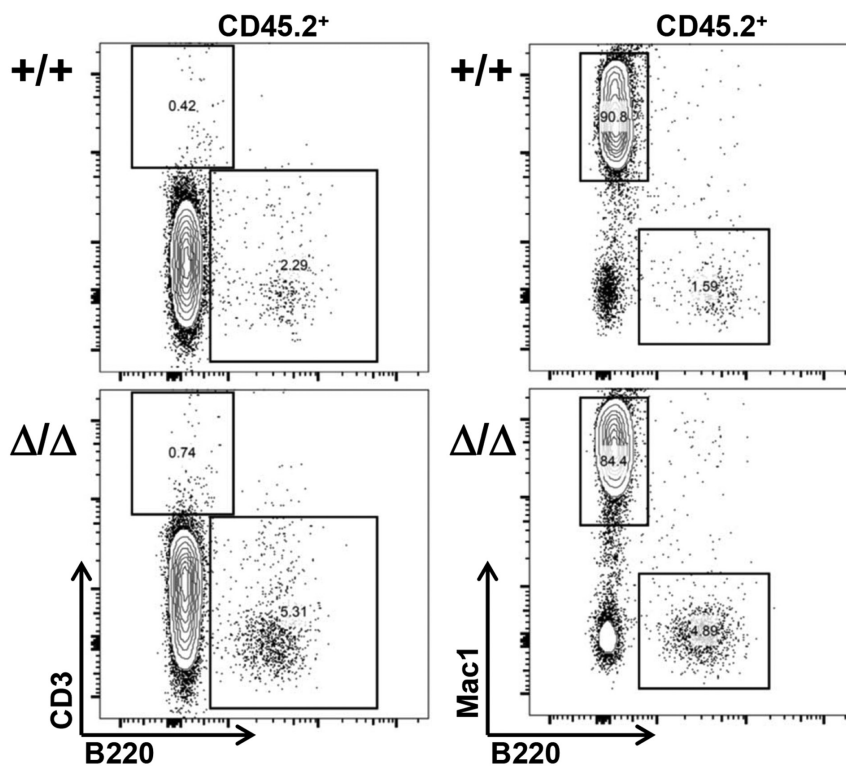


Figure S5. (A) WBC counts, LY, NEU and MO, and B platelet PLT measurements from recipient mice four weeks post-transplantation (n=5). N.s., not significant; WBC, white blood cells; LY, lymphocytes; NEU, neutrophils; MO, monocytes; PLT, platelet; HGB, hemoglobin.

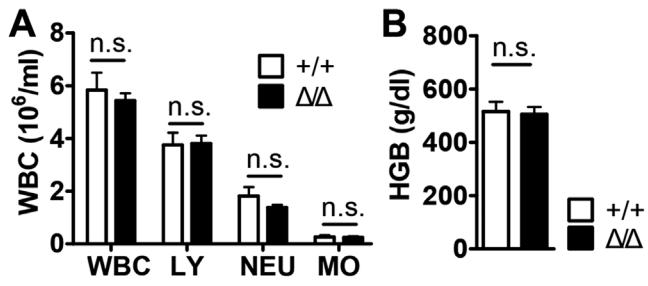


Table S1. Flow Cytometry Antibodies.

| Mature Cell Lineages | | | |
|-----------------------------|--|------------|----------|
| Antibody | Supplier | Cat. no. | Dilution |
| PE-Mac1 (M1/70) | BioLegend, Inc. | 101208 | 1:100 |
| FITC-B220R (RA3-6B2) | BD Biosciences | 553088 | 1:100 |
| APC-CD3 (145-2C11) | eBioscience; Thermo Fisher Scientific, Inc. | 17-0031-83 | 1:100 |
| For HSPC analysis | | | |
| Antibody | Supplier | Cat. no. | Dilution |
| PE-Cy5-CD3 (145-2C11) | BD Biosciences | 555276 | 1:100 |
| PE-Cy5-CD4 (RM4-5) | BD Biosciences | 553050 | 1:100 |
| PE-Cy5-CD8 (53-6.7) | BioLegend, Inc. | 100710 | 1:100 |
| PE-Cy5-CD19 (6D5) | BioLegend, Inc. | 115510 | 1:100 |
| PE-Cy5-B220R (RA3-6B2) | eBioscience | 15-0452-82 | 1:100 |
| PE-Cy5-Gr1 (RB6-8C5) | eBioscience | 15-5931-82 | 1:100 |
| PE-Cy5-Ter119 (TER119) | eBioscience | 15-5921-82 | 1:200 |
| PE-Sca1 (D7) | eBioscience | 12-5981-82 | 1:100 |
| APC-cKit (2B8) | BioLegend, Inc. | 105812 | 1:100 |
| PE-Cy7-CD150 (TC15-12F12.2) | BioLegend, Inc. | 115914 | 1:100 |
| APC-Cy7-CD48 (HM48-1) | BD Biosciences | 561242 | 1:500 |
| FITC-CD34 (RAM34) | eBioscience | 11-0341-85 | 1:50 |
| PE-Cy7-CD16/32 (93) | eBioscience | 25-0161-82 | 1:500 |
| APC-eFluor780-CD127 (A7R34) | eBioscience | 47-1271-82 | 1:100 |
| For CD45.2 Analysis | | | |
| Antibody | Supplier | Cat. no. | Dilution |
| FITC-CD45.2 (104) | BD Biosciences | 553772 | 1:100 |
| PE-Mac1 (M1/70) | Biolegend | 101208 | 1:100 |
| eFluor450- B220R (RA3-6B2) | eBioscience | 48-0452-82 | 1:100 |
| PE-Cy5-CD3 (145-2C11) | BD Biosciences | 555276 | 1:100 |

7-AAD staining was constructed using BrdU Kit according to the manufacturer's protocol (BD Biosciences). Briefly, 20 μ l 7AAD (BD) was added prior to FACS analysis. PE, R-phycoerythrin; FITC, fluorescein isothiocyanate; APC, allophycocyanin; Lin, lineage; Sca1, spinocerebellar ataxia type 1; cKit, mast/stem cell growth factor receptor kit; Mac1, macrophage-1 antigen; TER119, lymphocyte antigen 76.