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Title

Zeb1 Regulates the Function of Lympho-Myeloid Primed Progenitors after Transplantation

Authors

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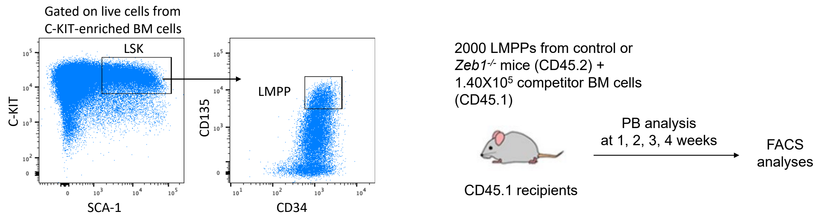
Introduction

Zeb1, a zinc finger E-box binding homeobox epithelial-mesenchymal (EMT) transcription factor, acts as a critical regulator of hematopoietic stem cell (HSC) self-renewal and multi-lineage differentiation. Whether *Zeb1* directly regulates the function of multi-potent progenitors primed for hematopoietic lineage commitment remains ill-defined.

Methodolgy

We used an inducible *Mx-1 Cre* conditional mouse model where *Zeb1* was genetically engineered to be deficient in the adult hematopoietic system (hereafter *Zeb1*$^{-/-}$).



Results

Loss of *Zeb1* Results in Blood Engraftment Defect after LMPP Transplantation

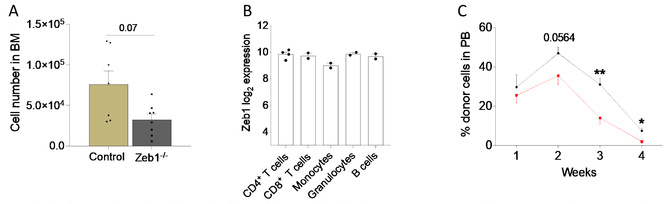


Figure 1. (A) Cell number of LMPPs (LSK CD34$^+$ CD135^{high}) in BM from control (n=7) and Zeb1$^{-/-}$ (n=8) mice 14 days after the last dose of pIpC from 4 independent experiments. (B) Zeb1 log2 expression data in subsets of mature blood cells. Data from BloodSpot. (C) The percentage of donor cells in PB at weeks 1, 2, 3, 4 post LMPP transplantation from control (n= 9-10, week 4 n=5) and *Zeb1*$^{-/-}$ (n= 9-10, week 4 n=5) mice from 2 independent experiments, except week 4 from one experiment. Error bars show mean ± SEM. Mann-Whitney U test was used to calculate significance as follows: *P < .05, **P < .01.

No T-Cell Engraftment from *Zeb1*$^{-/-}$ LMPPs after Transplantation due to Impact on T-cell Maturation

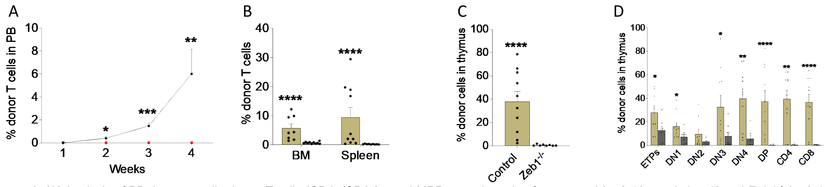


Figure 2. (A) Analysis of PB donor contribution to T cells (CD4$^+$ /CD8$^+$) post LMPP transplantation from control (n=9-10, week 4 n=5) and *Zeb1*$^{-/-}$ (n=9-10, week 4 n=5) mice from 2 independent experiments, except week 4 from one experiment. (B) Analysis of BM and spleen donor contribution to T cells (CD4$^+$ /CD8$^+$) 3-4 weeks post LMPP transplantation from control (n=9-10) and *Zeb1*$^{-/-}$ (n=9-10) mice from 2 independent experiments. The percentage of donor cells in thymus (C) and donor contribution to T cell populations in thymus (D) 3-4 weeks post LMPP transplantation from control (n=9-10) and *Zeb1*$^{-/-}$ (n=9-10) mice from 2 independent experiments. Error bars show mean ± SEM. Mann-Whitney U test was used to calculate significance as follows: *P < .05, **P < .01, ***P < .001, ****P <0.0001.

Reduced B-Cell and Monocyte/Macrophage Lineage Potential, but Unimpaired Granulocytic Differentiation from *Zeb1*$^{-/-}$ LMPPs after Transplantation

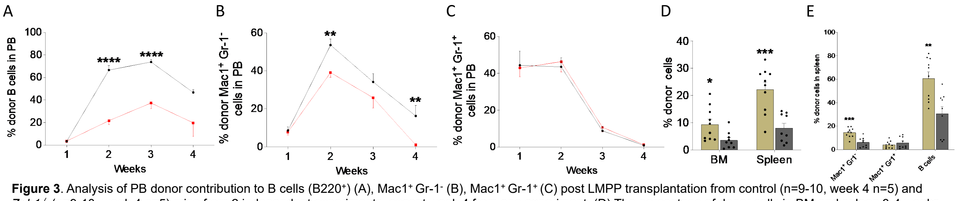


Figure 3. Analysis of PB donor contribution to B cells (B220$^+$) (A), Mac1$^+$ Gr-1$^-$ (B), Mac1$^+$ Gr-1$^+$ (C) post LMPP transplantation from control (n=9-10, week 4 n=5) and *Zeb1*$^{-/-}$ (n=9-10, week 4 n=5) mice from 2 independent experiments, except week 4 from one experiment. (D) The percentage of donor cells in BM and spleen 3-4 weeks post LMPP transplantation from control (n=9-10) and *Zeb1*$^{-/-}$ (n=9-10) mice from 2 independent experiments. (E) Percentage of donor cells in spleen and donor contribution to B cells (B220$^+$), Mac1$^+$ Gr-1$^-$, and Mac1$^+$ Gr-1$^+$ post LMPP transplantation from control (n=9-10) and *Zeb1*$^{-/-}$ (n=9-10) mice from 2 independent experiments. Error bars show mean ± SEM. Mann-Whitney U test was used to calculate significance as follows: *P < .05, **P < .01, ***P < .001, ****P <0.0001.

Conclusion

Zeb1 mediates LMPP differentiation to T-cell, B-cell, and monocyte/macrophage lineages but is expendable for the regulation of granulocyte differentiation in vivo.

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