

**The osteoprogenitor-specific loss of ephrinB1 results in an osteoporotic phenotype affecting the balance between bone formation and resorption**

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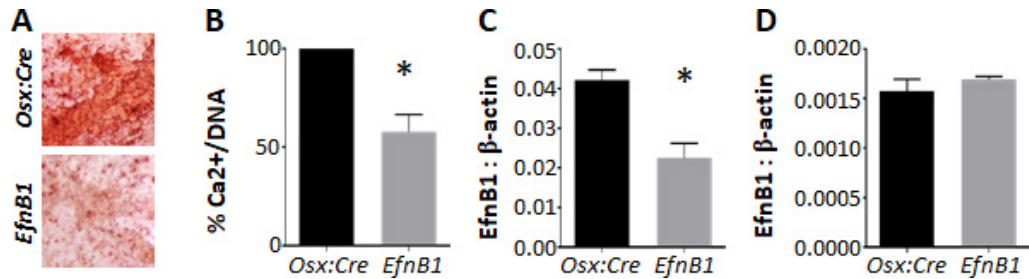
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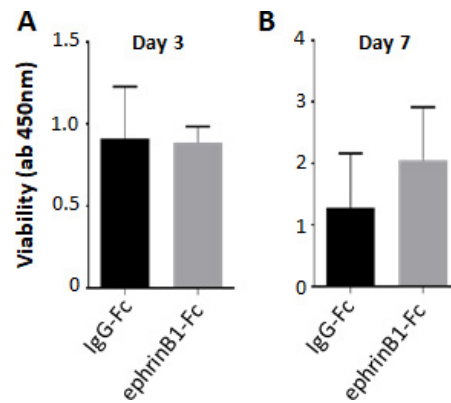
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**Running Title: loss of ephrinB1 results in an osteoporotic phenotype**

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**Supplementary Figure 1. Loss of *EfnB1* in osteoprogenitors mediated through the osterix promoter influences osteoblast in vitro.** (A-C) *Osx:Cre* (*Osx*) and *EfnB1*<sub>OB</sub><sup>-/-</sup> (*EfnB1*) cells were cultured under osteogenic differentiation conditions for 21 days. Cultures were either (A) stained with Alizarin Red, (B) quantitated for calcium ( $\text{Ca}^{2+}$ ) levels and represented as a percentage of  $\text{Ca}^{2+}$ /DNA relative to the *Osx:Cre* control, or (C) isolated for PCR analysis of *ephrinB1* gene expression. (D) *Osx:Cre* and *EfnB1*<sub>OB</sub><sup>-/-</sup> bone marrow cells were cultured under osteoclast conditions. Samples were isolated for PCR analysis of *ephrinB1* gene expression. (All data presented, n = 2-3 mice/strain, \* p<0.05, Student t-test).



**Supplementary Figure 2. Soluble ephrinB1 does not influence osteoclast precursor viability or metabolic function.** (A-B) Human PBMNC viability and metabolic function was assessed by WST-1 analysis (absorbance 450nm) on (A) Day 3 and (B) Day 7 following osteoclast differentiation, in the presence of human IgG-Fc or ephrinB1-Fc (n = 2 human donors, Student t-test).