

Supplemental Materiel

Supplemental Table 1 Primer Sequence:

Name	Sequence
Angiopoietin	F: 5'CTCGTCAGACATTCATCATCCAG3' R: 5'CACCTTCTTTAGTGCAAAGGCT3'
SFRP2	F: 5'CCCTCATGAGCTCTGACCAT3' R: 5'TATTTGAGGGCATCATGCAA3'
SFRP1	F: 5'CGAGTTTGCCTGAGGATGA3' R: 5'CGTTCTTCAGGAACAGCACAA3'
SPP1 (osteopontin)	F: 5'TCCCTCGATGTCATCCCTGTTG3' R: 5'GGCACTCTCCTGGCTCTCTTTG3'
WNT3a	F: 5'TTTGGAGGAATGGTCTCTCG3' R: 5'ACCACCAGCAGGTCTTCACT3'
CXCL12	F: 5'TGCATCAGTGACGGTAAACCA3' R: 5'CACAGTTTGGAGTGTTGAGGAT3'
CDH2	F: 5'TCCACCTCGCTGTAAAAATGG3' R: 5'CATGCAAAAAACCTGAATCCAA3'
BMP4	F: 5'TAAGAAGTCCGTCGCCATT3' R: 5'GGCCACAATCCAATCATTCC3'
MMN (nestin)	F: 5'GCCTCAGACATAGGTGGGATG3' R: 5'CCCCTTGCCTAATACCCTTGA3'
LEF1	F: 5'GCCACCGATGAGATGATCCC3' R: 5'TTGATGTCGGCTAAGTCGCC3'
PPAR-GAMMA	F: 5'ACCACTCGCATTCTTTGAC3' R: 5'TGGGTCAGCTCTTGTGAATG3'
C/EBP ALPHA	F: 5'CATCAGCGCCATCATCGACC3' R: 5'CCAGGAAGTTCGTCGTTGAAG3'
Alkaline phosphatase	F: 5'GAGCGTCATCCCAGTGGAG3' R: 5'TAGCGGTTACTGTAGACACCC3'
RUNX2	F: 5'AGAGTCAGATTACAGATCCCAGG3' R: 5'AGGAGGGGTAAGACTGGTCATA3'
Osteocalcin	F: 5'CTGACCTCACAGATCCCAAGC3' R: 5'TGGTCTGATAGCTCGTCACAAG3'
SHH	F: 5'AAAGCTGACCCCTTTAGCCTA3' R: 5'TTCGGAGTTTCTTGTGATCTTCC3'
Beta actin	F: 5'GCTCTTTTCCAGCCTTCCTT3' R: 5'CTTCTGCATCCTGTCAGCAA3'

Supplemental Table 2: p190-B-deficiency causes erythropoiesis defects at e14.5

	P1	P2	P3	P4	P5
frequency					
+/+	2.5±0.3	4.12±0.72	57±7.7	13.9±4.52	1.35±0.34
+/-	2.78±0.5	3.9±0.86	58±5.7	14±5.39	1.29±0.65
-/-	3.17±0.35*	5.24±0.85*	54±5.7	8.5±3.12*	0.55±0.3*
total cell number per liver					
+/+	0.33±0.039	0.54±0.095	7.56±1.01	1.87±0.6	0.18±0.045
+/-	0.37±0.062	0.52±0.11	7.68±0.75	1.86±0.71	0.17±0.086
-/-	0.17±0.019*	0.29±0.046*	2.97±0.33*	1.47±0.17	0.03±0.016*

P1: Ter119^{low}CD71^{low}; P2: Ter119^{low}CD71^{high}; P3: Ter119^{high}CD71^{high}; P4: Ter119^{high}CD71^{Int.}; P5: Ter119^{high}CD71^{low}, profile of various erythropoietic population according to (REF). * p<0.05, n=7-9

Note that the frequency of P1 and P2 population were increased in p190-B^{-/-} fetal liver whereas the frequency of P4 and P5 population were decreased; overall the total numbers of erythroid cells at various stage of differentiation were decreased in p190-B^{-/-} fetal livers compared to WT

Supplemental Table 3: Blood parameters of the 10% of p190-B haploinsufficient adult animals that live shortly

	+/+	+/-
WBC ($10^3/\mu\text{l}$)	9.398 \pm 1.098	9.142 \pm 2.062
Gr1/Mac1 (%)	15.7 \pm 5.9	36.1 \pm 24.7*
B220 (%)	50.5 \pm 8.4	30.4 \pm 15.5*
T cells (%)	22.5 \pm 3.1	24 \pm 9.5
RBC ($10^6/\mu\text{l}$)	8.859 \pm 0.4366	10.15 \pm 0.534
Hb (g/L)	13.54 \pm 0.65	15.39 \pm 0.8872
Platelets ($10^6/\mu\text{l}$)	731.8 \pm 63.14	758.7 \pm 62.21

WBC: white blood cell, RBC: red blood cells, Hb: hemoglobin,
Gr1/Mac1: granulocytes/monocytes, B220: B lymphocytes, T cells: T lymphocytes
* p<0.05, n=8

Supplemental figure legends

Supplemental Figure 1.

(A) Weight and **(B)** cellularity (cell number per liver) of e14.5 fetal livers. **(C)** Fetal liver cell size assessed by forward scatter parameter measurement using flow cytometry.

Supplemental Figure 2.

Representative flow cytometry chart of erythroid differentiation profile assessed by Ter119 and CD71 expression from BM cells of WT recipients reconstituted with WT or p190-B^{-/-} FL cells.

Supplemental Figure 3.

Transplantation of WT cells in WT and p190-B heterozygotes mice that live normally.

(A) Total number of cells per BM (right panel) and spleen (left panel). **(B)** Frequency of LSK-SLAM, LSK and LK progenitors, **(C)** Frequency of B cell (B220+) and granulomonocytic cells (Gr-1+CD11b+) in BM. (mean±SD, n=8-10 from 2 independent experiments).

Supplemental Figure 4

Analyses of cultured-derived stromal cells **(A)** Representative pictures of cultured-derived stromal cells. **(B)** Representative flow analyses of expression of markers of MSCs. **(C)** qPCR analysis of the mRNA expression of putative MSC markers in the stromal cells. Data are expressed as log₂ fold relative to average of WT samples set to 0. n=3-4 samples per group, each triangle represents one independent sample.

Supplemental Figure 5

Flow chart of the analysis of immunophenotypic markers of stromal cells in the fetal liver.

Supplemental Figure 6

(A) Number of CFU-F per e14.5 fetal livers (mean±SD, n=4). **(B)** Immunophenotypic characterization of e14.5 fetal liver stromal cells, (mean±SD, n=8).

Supplemental Figure 7

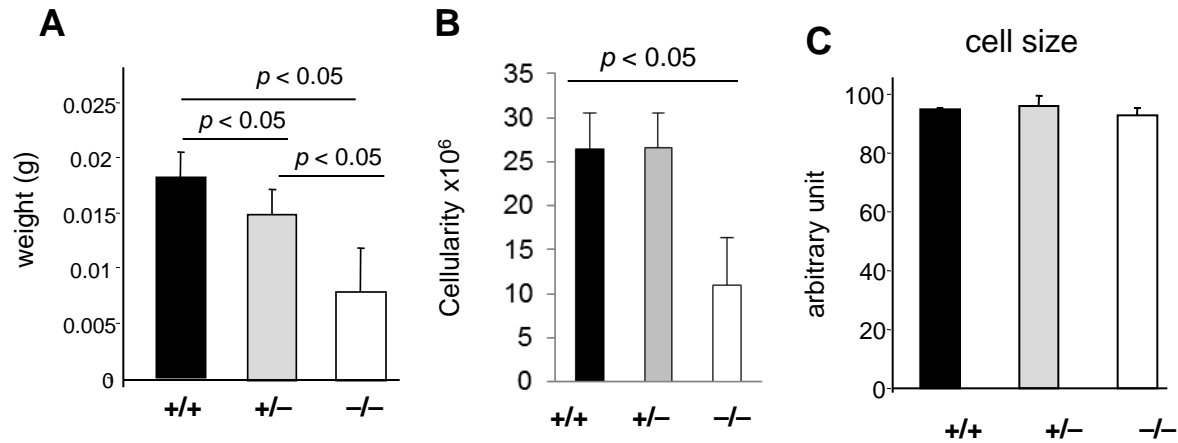
Masson's trichrome staining adult femur from of WT and p190-B heterozygotes that live shortly, enlarged pictures (representative of n=3). Note the decrease in collagen deposition indicated by lighter blue intensity in p190-B heterozygotes.

Supplemental Figure 8

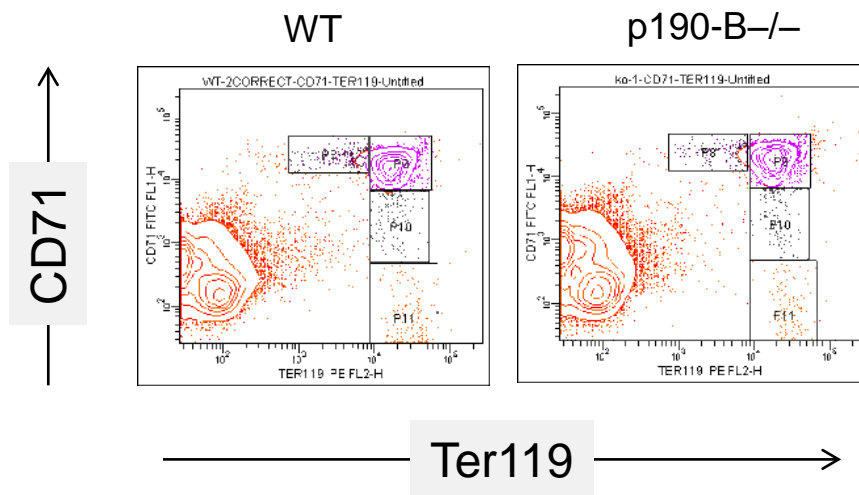
qPCR analysis of mRNA expression of the indicated genes. Data are expressed as log₂ fold relative to average of WT samples set to 0. *p<0.05 Mann-Whitney test, n=3-4 samples per group, each triangle represents one independent sample

Supplemental Figure 9

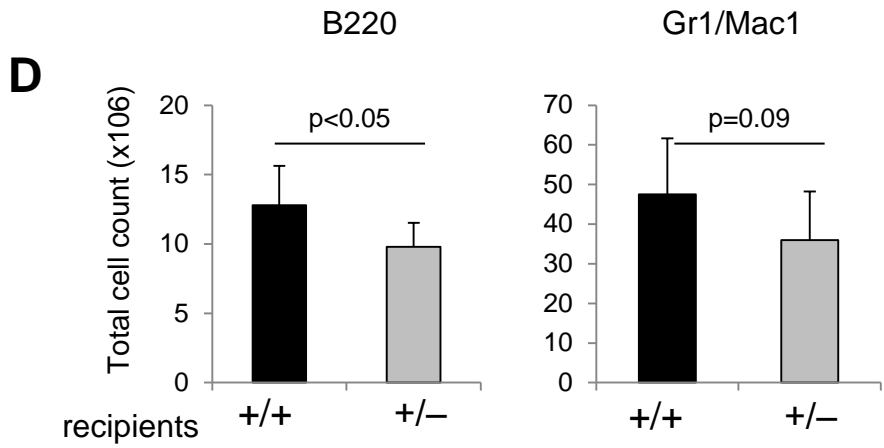
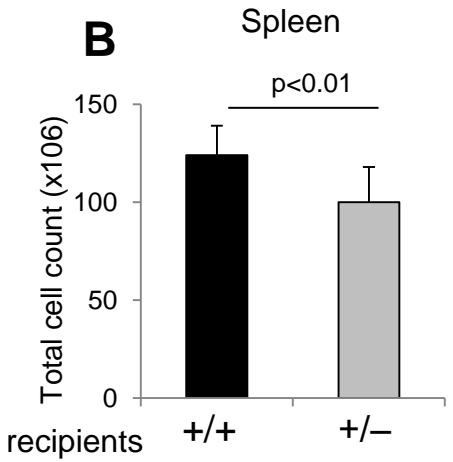
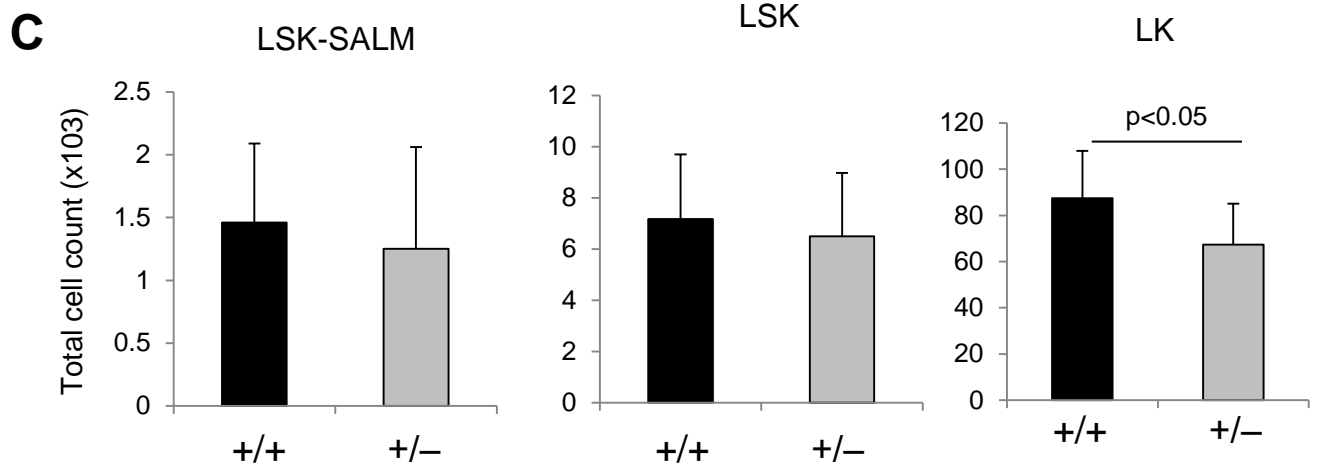
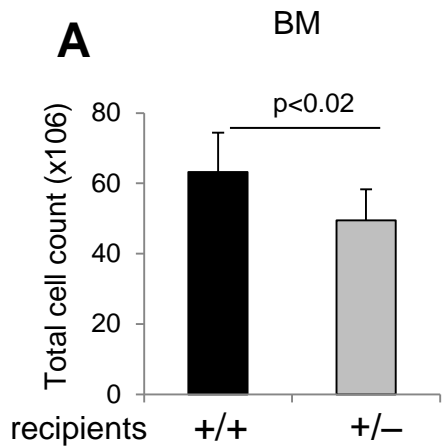
Alizarin red and alcian blue staining of skeleton at e18.5. Arrow points to visible skeletal defects, which were observed in each of the p190-B^{-/-} embryos examined (n=5 per genotypes).



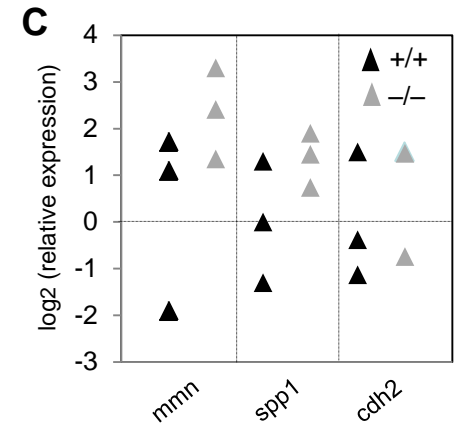
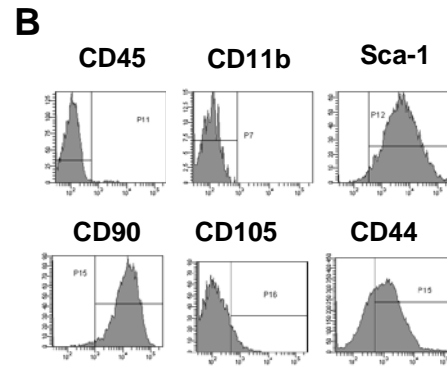
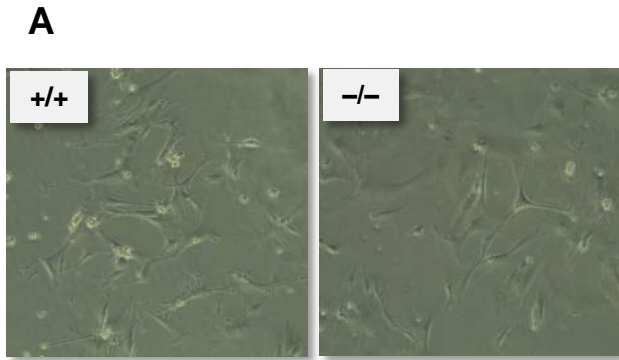
Supplemental figure 1



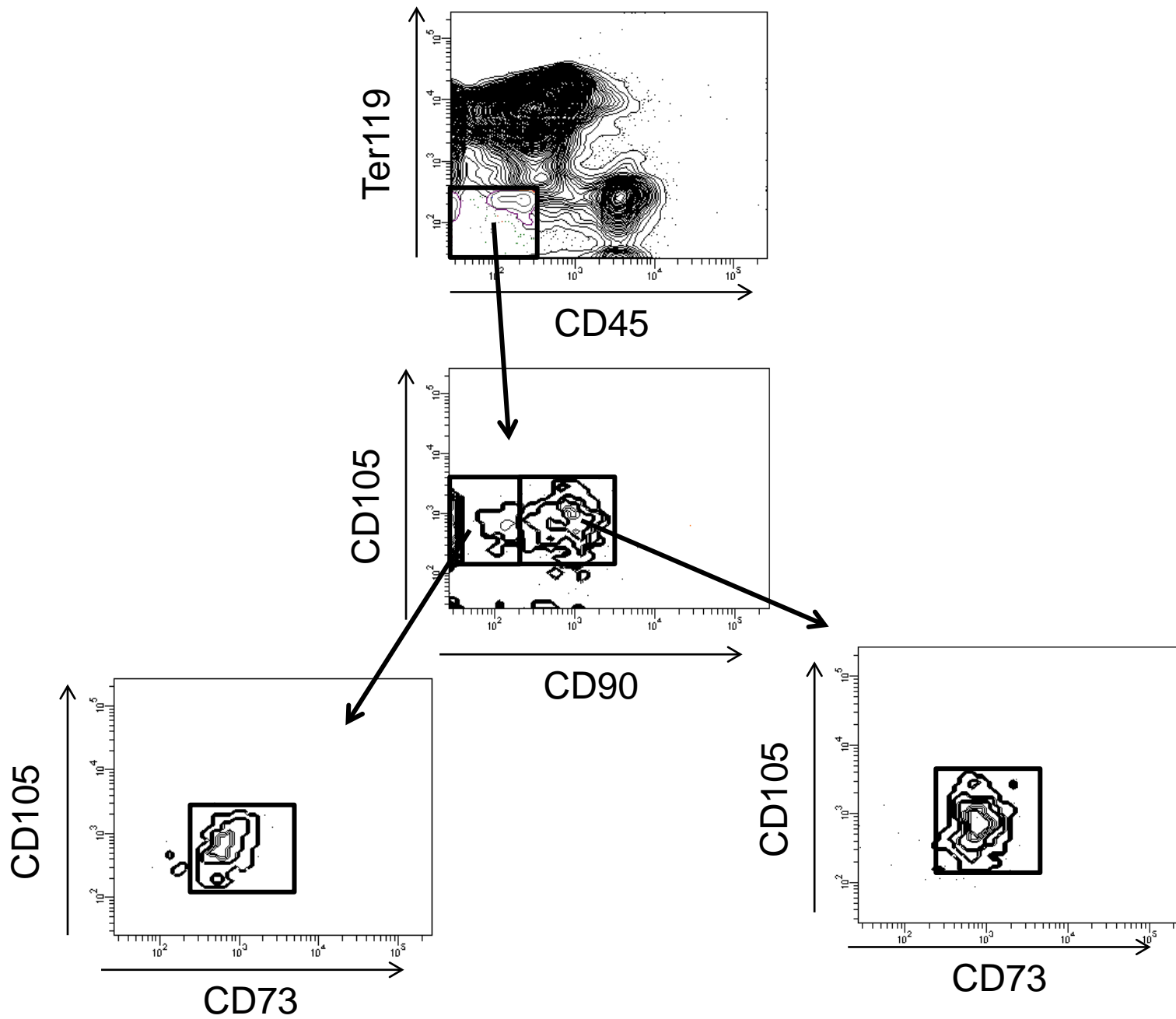
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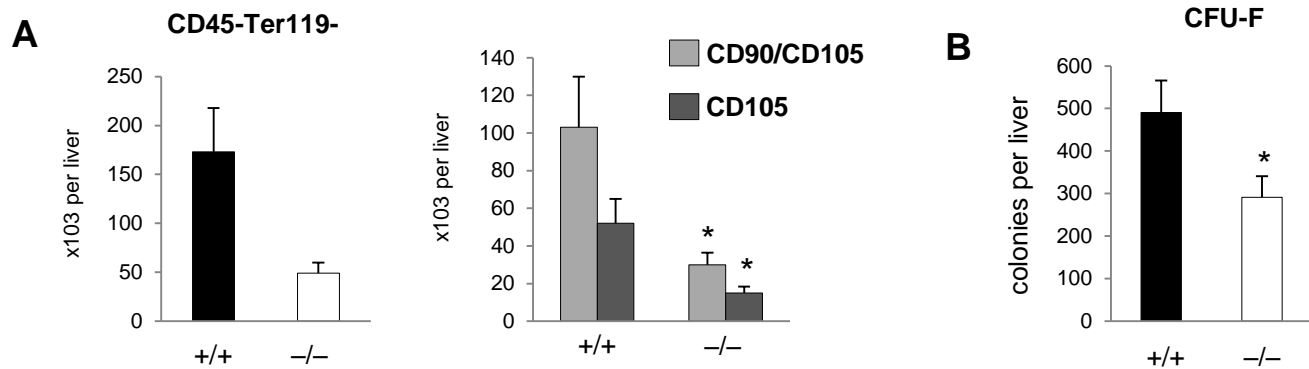
Supplemental Figure 3



Supplemental figure 4



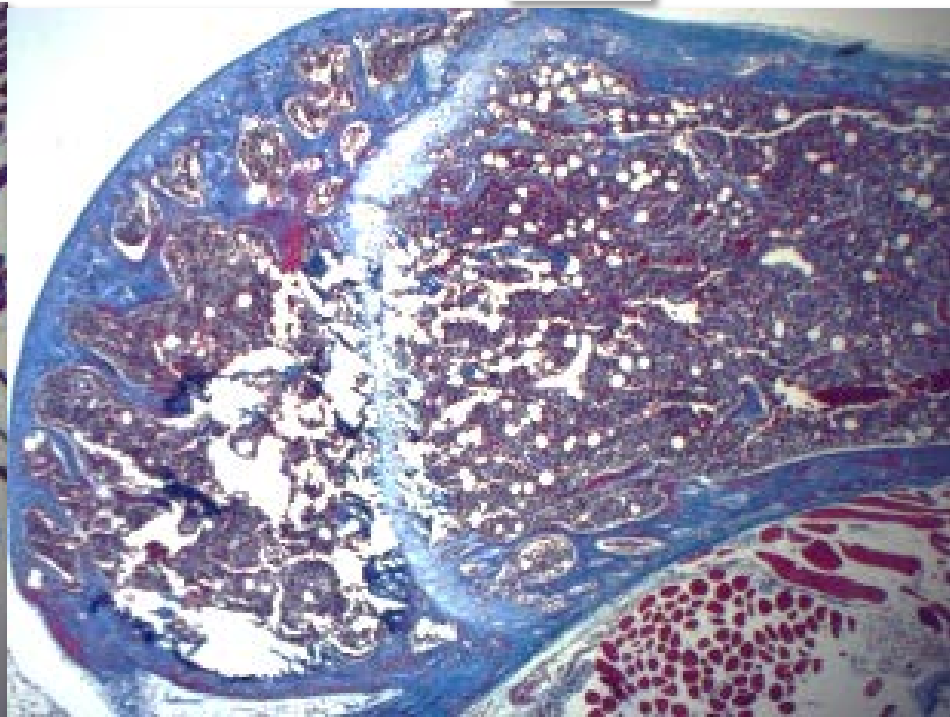
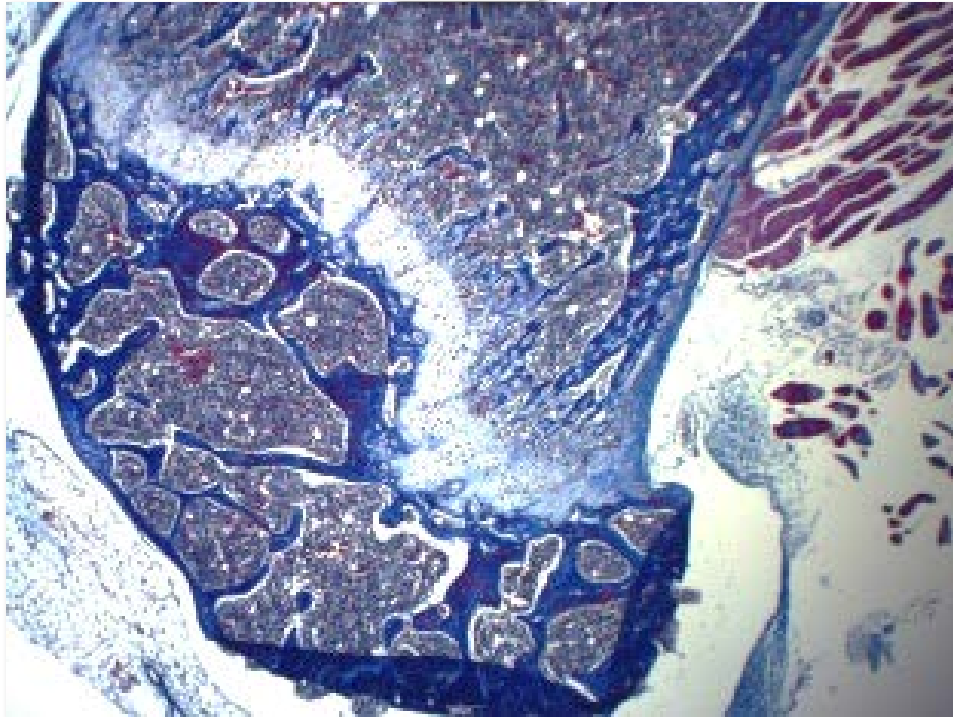
Supplemental Figure 5



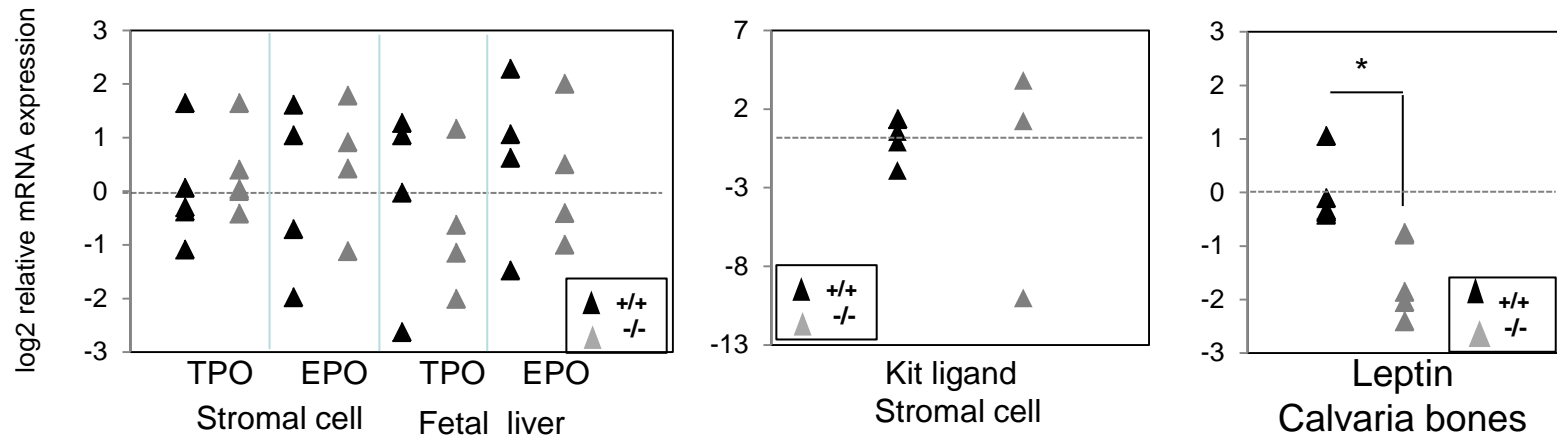
Supplemental Figure 6

+/+

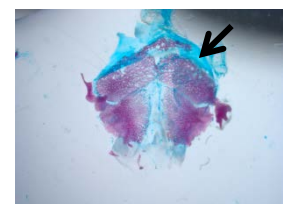
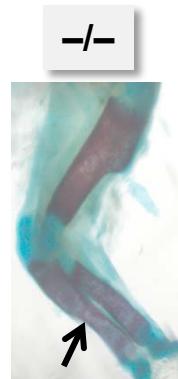
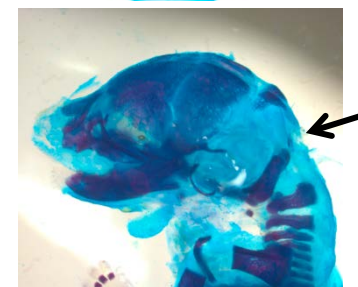
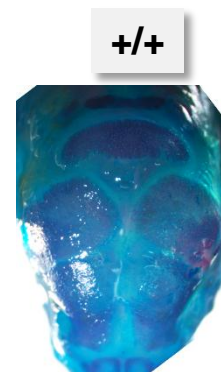
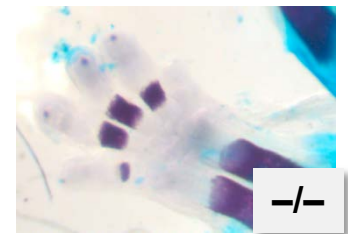
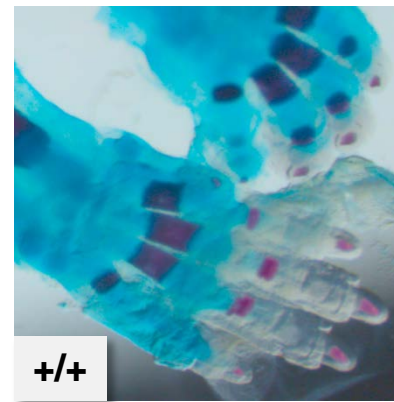
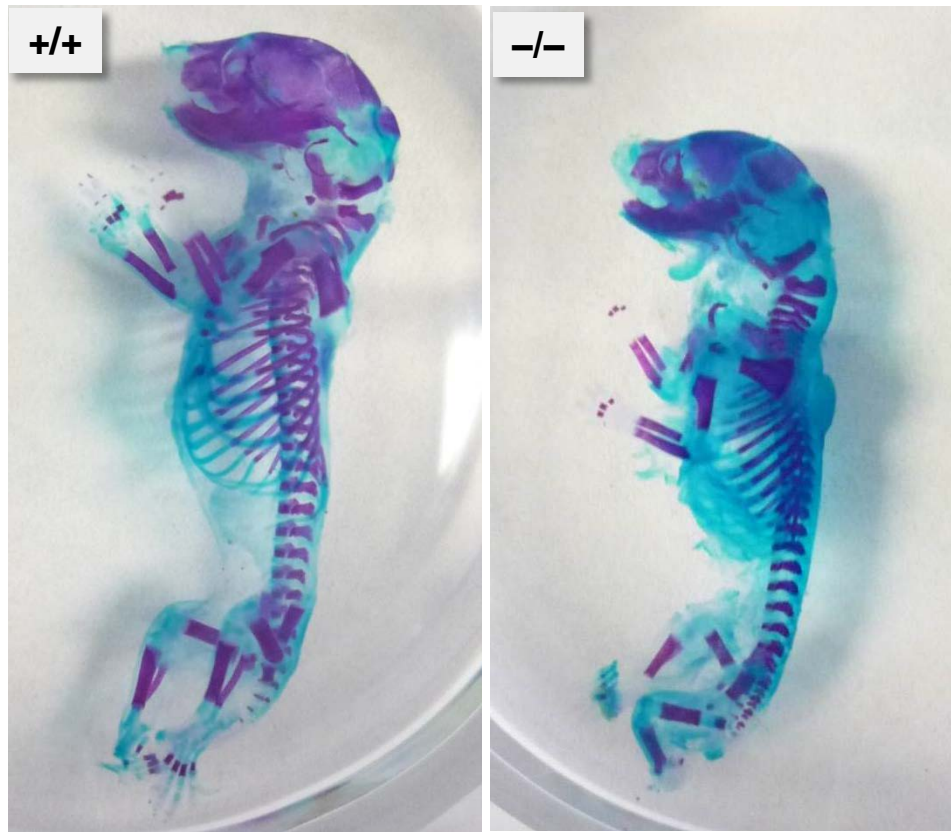
+/-



Supplemental Figure 7



Supplemental Figure 8



Supplemental figure 9