

Supplementary Figure 1: Mutant mice (Selm^{-/-}) were generated from C57BL/6 embryonic stem cell clones 11537A-A8 and 11537A-B11 (referred to as AA8 and AB11) by injection into blastocysts from C57BL/6 mice with genetically deleted Selm obtained from the knockout mouse project (KOMP) repository (an NCRR-NIH-supported strain suppository). ES-cells were created by VelociGene from funds provided by the trans-NIH KOMP. Subsequent intercrosses of heterozygous animals generated two congenic strains with a C57BL/6 genetic background. (A) The Selm ORF was deleted and replaced by a ZEN-Ub1 cassette harbouring the reporter and neo resistance gene. Genotyping primers are indicated by arrows. lacZ: β-galactosidase coding sequence from E. coli lacZ gene, hUbCpro: promoter from the human Ubiquitin C gene, neor: coding sequence for neomycin phosphotransferase. (B) Southern blot analysis of genomic DNA from the kidney of wild type (+/+) and Selm- mice (AA8/AB11) using a probe against a 493 bp fragment of the LacZ gene from the knockout cassette (5'-gccaggcacagatgggtaccg-3' and 5'-taaccgacccagcgcccgtt-3'). (C) Genotypes were determined by multiplex PCR using different primer pairs for wild type (-/-; 583 bp) and knockout (-/-; 392 bp) mice (wild type: 5'- tcagccaaatgacccgggacg-3' and 5'- ctgccccgtctgtcaaaacacc-3'; knockout: 5'- tttgggtgcagcctgcggaa-3' and 5'- ttctccgtgggaacaaacggcg-3'). +/-, heterozygous mice. (D) RNA in situ hybridization on hindlimb sections of postnatal day 4 wild type (+/+) and Selm^{-/-} (-/-) mice. 5 μm paraffin sections were hybridized with antisense riboprobes against Selm; scale bar: 0.5 mm. (E) Selm expression of wild type (+/+) and Selm-1- (-/-) P4 calvarial osteoblasts relative to Actb expression (mean + S.D., n = 4).