

Figure S1

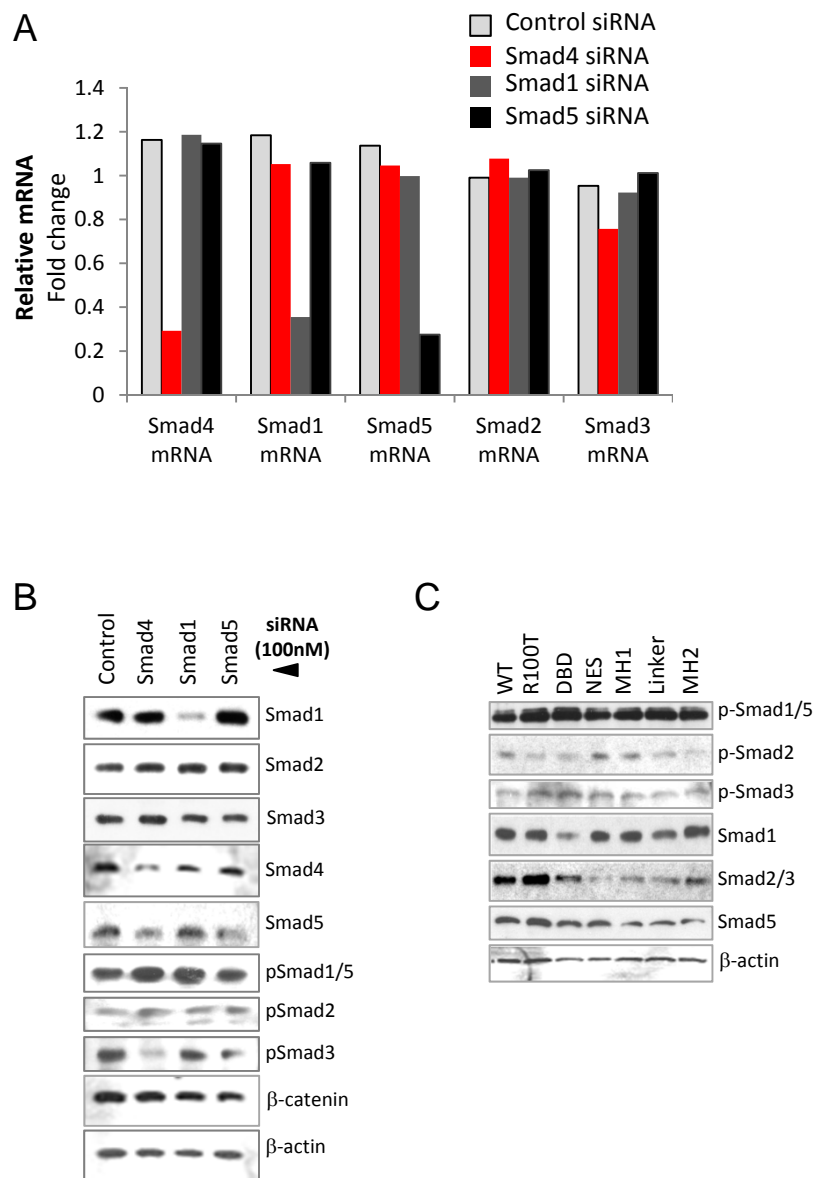


Figure S1 (A) QPCR on MC3T3 cells transfected with indicated siRNA. Data represent the average of 2 experiments. **(B)** Immunoblot of whole cell lysates transfected with indicated siRNA. **(B)** Immunoblot on MC3T3 cells transfected with HA-Smad4 mutants.

Figure S2

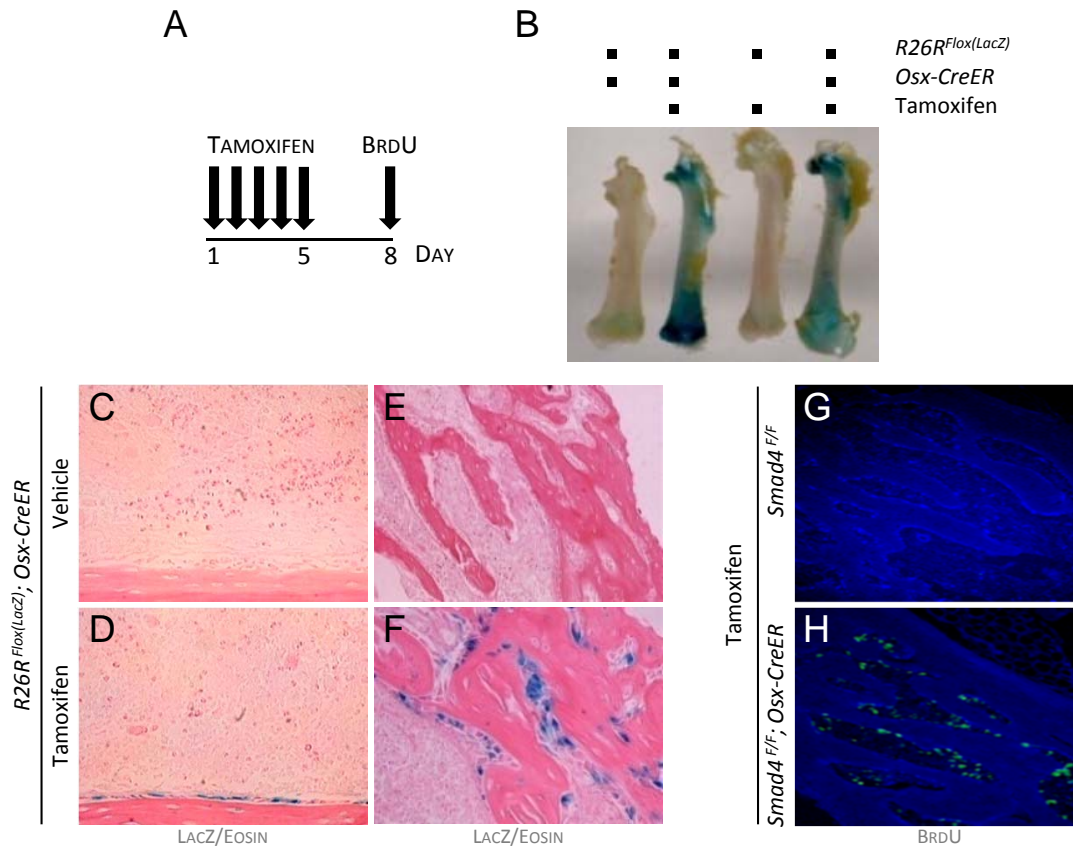


Figure S2 Tamoxifen-dependent ablation of *Smad4* in *Osx*⁺ cells of adult mice increases proliferation of cells adjacent to trabecular bone surfaces. **(A)** CreER activity was induced in *Osx-CreER* mice using 5 consecutive daily injections of tamoxifen (100 mg/kg i.p). Mitotic cells were labeled with a single dose of BrdU 2 hours prior to sacrifice on day 8. **(B)** Intact femurs were subjected to LacZ staining, **(C-F)** histologically sectioned, and then counterstained with Eosin to evaluate LacZ⁺ cells in bone by bright field microscopy. **(C-D)** 20X images of the cortex in the mid-diaphysis and **(E-F)** 40X images of trabecular bone in the distal femur showing LacZ⁺ cells on bone surfaces and embedded in bone matrix. **(G-H)** *Smad4*^{F/F} and *Smad4*^{F/F}; *Osx-CreER* mice were treated with tamoxifen and BrdU as indicated in **(A)**. Mitotic cells were visualized in histologic sections of the trochanter by BrdU immunofluorescence and DAPI.