Supporting Information

Wei et al. 10.1073/pnas.1200797109

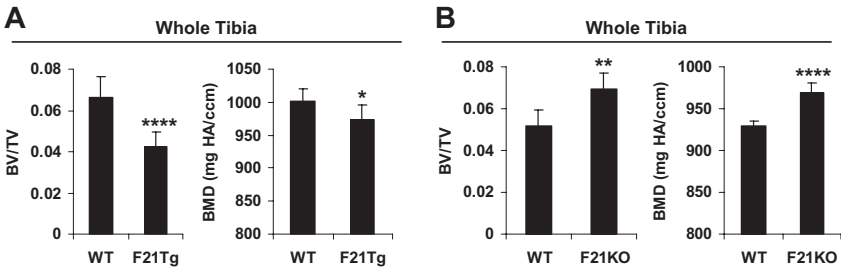

Fig. S1. μCT analyses of the whole tibiae from FGF21-Tg and FGF21-KO mice. (*A*) The whole tibiae from FGF21-Tg mice showed decreased bone volume and mineral density (6-mo-old male, $n = 7$). (*B*) The whole tibiae from FGF21-KO mice showed increased bone volume and mineral density (4-mo-old male, $n = 4$). $^{*}P < 0.05$; $^{**}P < 0.01$; $^{****}P < 0.001$.

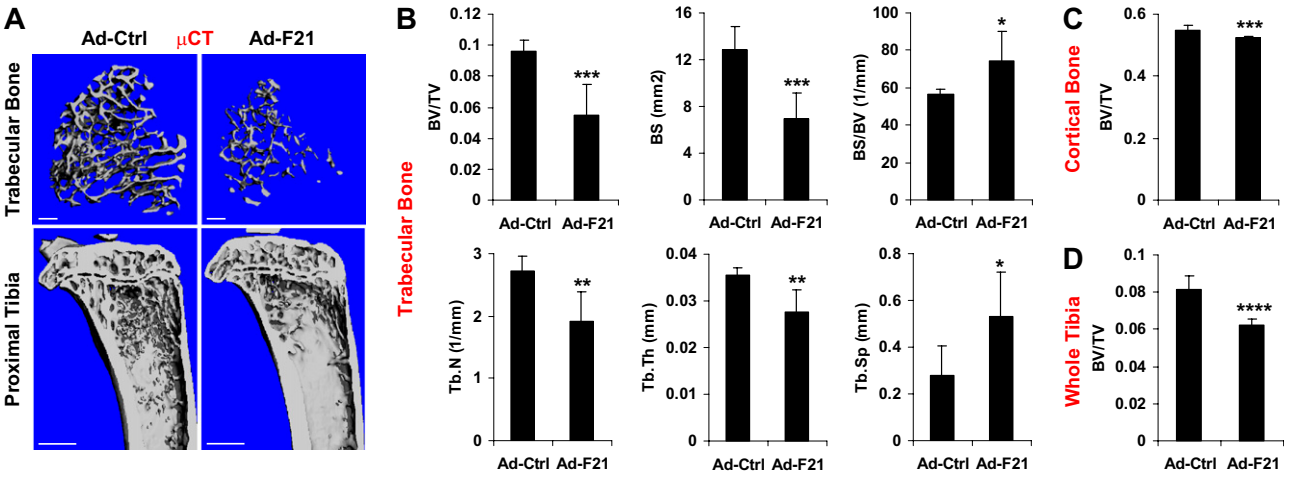

Fig. S2. Adenovirus-mediated FGF21 overexpression causes bone loss in obese/diabetic mice. Mice were fed a high-fat diet for 3 mo, injected with 7.5×10^9 adenoviral particles per gram of body weight via jugular vein, and dissected 2 wk later (6-mo-old at end point, male, $n = 6$). Tibiae from mice infected with control virus (Ad-ctrl) or FGF21 virus (Ad-F21) were scanned and analyzed by a Scanco μCT35 instrument. (*A*) Representative images of the trabecular bone of the tibial metaphysis (*Upper*) and the entire proximal tibia (*Lower*). (Scale bars: *Upper*, 10 μm; *Lower*, 1 mm). (*B*) Quantification of trabecular bone volume and architecture. (*C*) BV/TV of the cortical bone. (*D*) BV/TV of the whole tibiae. $^{*}P < 0.05$; $^{**}P < 0.01$; $^{***}P < 0.005$; $^{****}P < 0.001$.

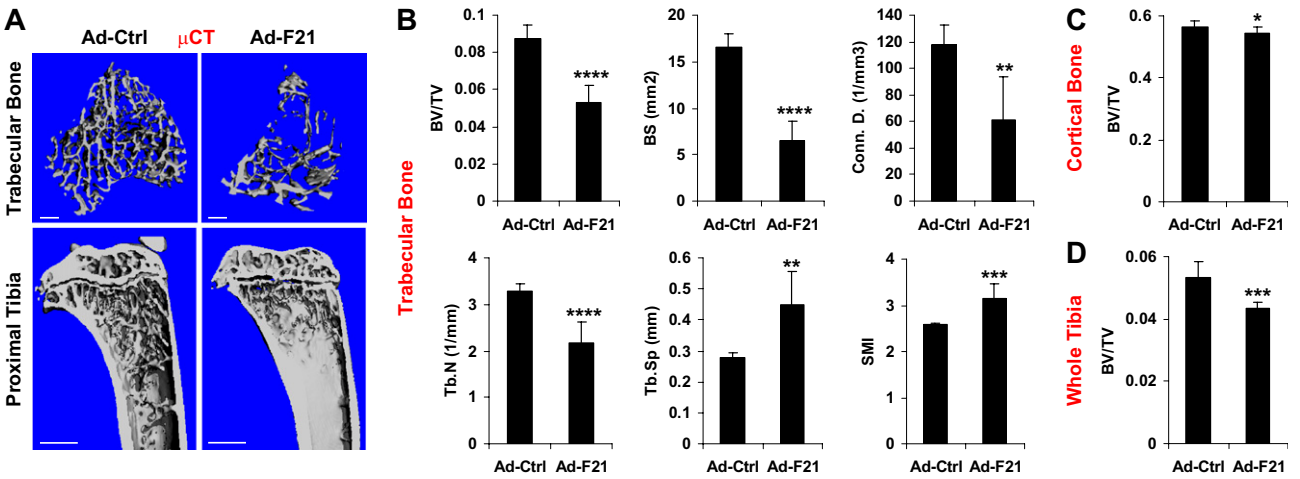

Fig. S3. Adenovirus-mediated FGF21 over-expression causes bone loss in lean mice. Mice were fed a chow diet, injected with 7.5×10^9 adenoviral particles per gram of body weight via jugular vein, and dissected 1 mo later (3-mo-old at end point, male, $n = 5$). Tibiae from mice infected with control virus (Ad-ctrl) or FGF21 virus (Ad-F21) were scanned and analyzed by a Scanco μCT35 instrument. (*A*) Representative images of the trabecular bone of the tibial metaphysis (*Upper*) and the entire proximal tibia (*Lower*). (Scale bars: *Upper*, 10 μm; *Lower*, 1 mm). (*B*) Quantification of trabecular bone volume and architecture. SMI, structure model index (a higher SMI indicates weaker bone). (*C*) BV/TV of the cortical bone. (*D*) BV/TV of the whole tibiae. $*P < 0.05$; $**P < 0.01$; $***P < 0.005$; $****P < 0.001$.

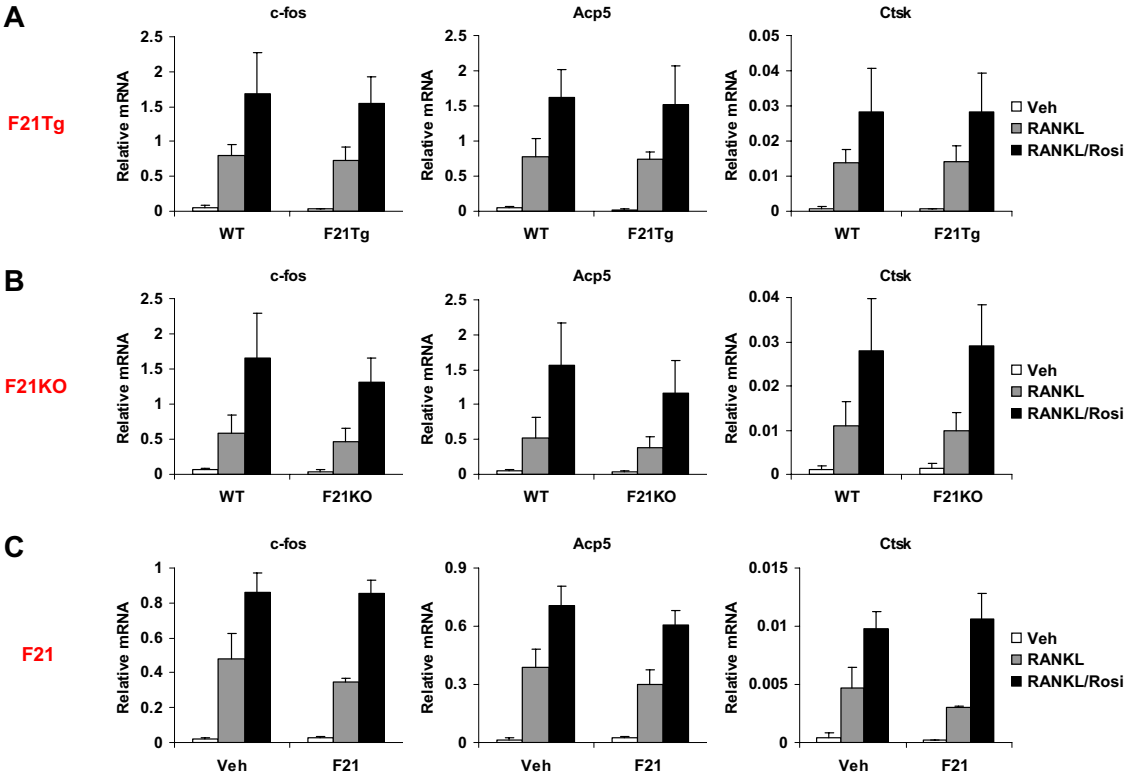

Fig. S4. FGF21 does not directly regulate osteoclast differentiation ex vivo. Bone marrow cells from WT, FGF21-Tg, or -KO mice were differentiated ex vivo with MCSF and RANKL, with or without rosiglitazone (Rosi), in the absence or presence of recombinant FGF21 (F21). Osteoclast differentiation was quantified by marker gene expression using qRT-PCR. Acp5, acid phosphatase 5, tartrate resistant, i.e., TRAP; Ctsk, cathepsin K. (*A*) The bone marrow cells of FGF21-Tg mice had unaltered osteoclastogenic potential ex vivo. (*B*) The bone marrow cells of FGF21-KO mice had unaltered osteoclastogenic potential ex vivo. (*C*) FGF21 treatment alone had no effect on osteoclast differentiation of WT bone marrow cells ex vivo.