Fig. S1. Morphological changes of apoptotic muscle fibroblasts induced by myostatin inhibitor. Primary muscle fibroblasts from limb muscle of mdx mice were treated with PBS, ActRIIB.mFc, etoposide (ET) or cycloheximide (CHX) for 12 hours. Apoptotic fibroblasts were identified by immunostaining for expression of cleaved caspase-3. Cellular morphology was demonstrated by immunostaining for α-smooth muscle actin (SMA). Cellular nuclei were stained with 4′,6-diamidino-2-phenylindole (DAPI). The histogram depicts the percent of apoptotic fibroblasts in each treatment group. **P<0.01
Fig. S2. Inhibition of myostatin induces muscle fibroblasts to undergo apoptosis in mdx mice. Immunohistochemistry on cross-sections of triceps muscle from mdx mice treated with ActRIIB.mFc for 6 weeks (A): Apoptotic cells detected with TdT-mediated dUTP nick end labeling assay (TUNEL) co-localize with cleaved caspase-3 positive cells. (B-D): Apoptotic cells detected by cleaved caspase-3 expression (B) and TUNEL assay (C and D) colocalize with muscle fibroblasts detected by fibroblast markers vimentin (B), ER-TR7(C) and collagen I (D). Scale bar: 40\(\mu\)M.
Fig. S3. Magnetic resonance imaging (MRI) of muscle fibrosis. (A) T1-weighted MRI of calf muscles in C57BL/6 and mdx mice before and after injection of paramagnetic contrast agent gadolinium. (B) By subtracting post-contrast T1-weighted MR images from pre-contrast T1-weighted MR images, regions with signal enhancement in the hindlimb muscles were outlined and T2 values in these regions were obtained from coregistered T2 maps. (C) MRI of calf muscle in two-year-old mdx mice before and after treatment with PBS for 8 weeks. Histograms show no significant changes in calf muscle volume and T2 values in PBS treated animals.