

Figure S1. Transfection efficiency of si-FoxO6 in C₂C₁₂ cells. C₂C₁₂ cells were transfected with si-FoxO6-1, si-FoxO6-2, si-FoxO6-3 or si-NC. At 48 h post-transfection, transfection efficiencies were determined via (A) reverse transcription-quantitative PCR and (B) western blotting. β -actin was used as the loading control. **P<0.01, ***P<0.001 and ****P<0.0001. si, small interfering RNA; FoxO6, forkhead box O6; NC, negative control.

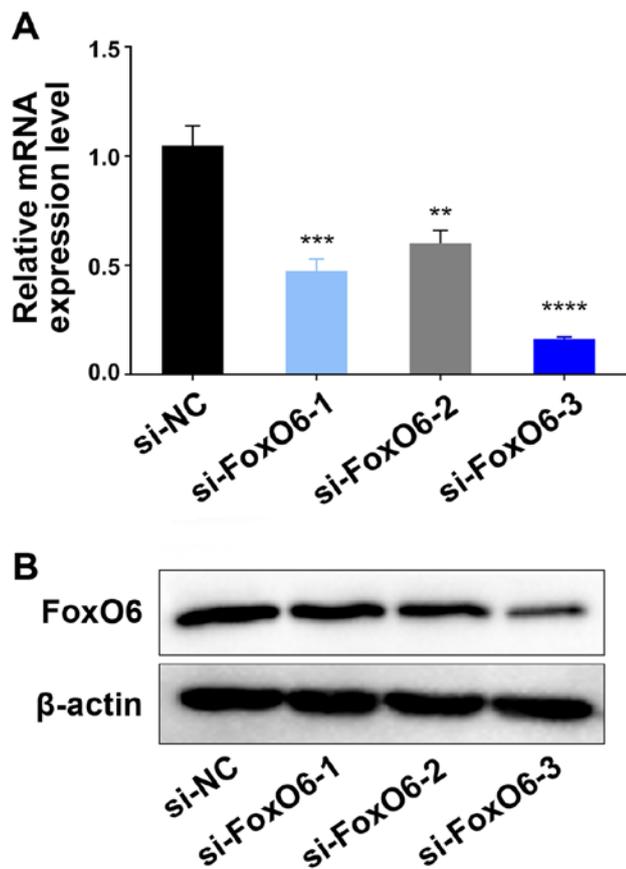


Figure S2. Effect of FoxO6 knockdown on MyHC expression in C₂C₁₂ myotubes. At 48 h post-transfection, MyHC expression was detected via immunofluorescence staining analysis. FoxO6, forkhead box O6; MyHC, myosin heavy chain; si, small interfering RNA; NC, negative control.

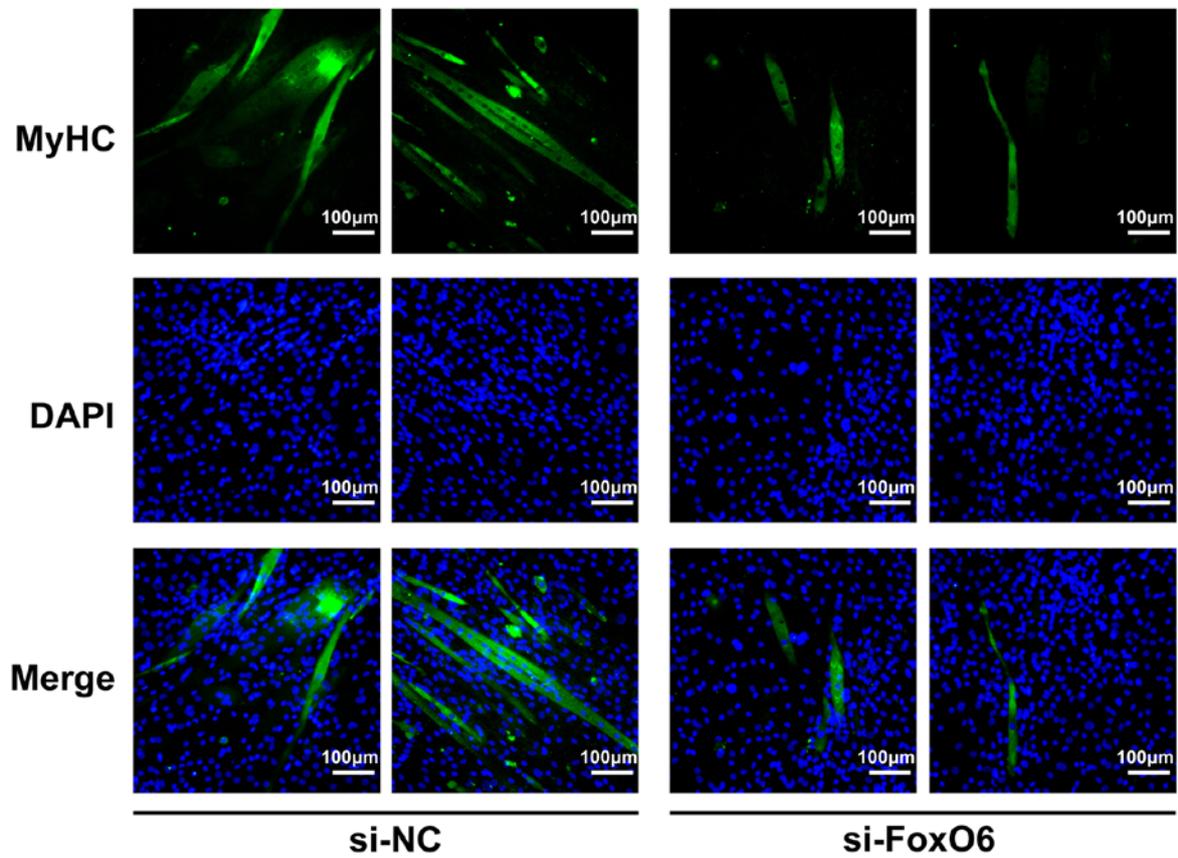


Figure S3. Knockdown efficiency of AAV9-shFoxO6 in the skeletal muscles of mice. Knockdown efficiency was determined via reverse transcription-quantitative PCR, and each sample was examined in duplicate. Data are presented as the mean \pm SEM. Compared with the mean of the five mice in the control group, the result demonstrated that FoxO6 mRNA expression levels were 65% decreased in the AAV9-shFoxO6 group. *** $P < 0.001$. AAV9, adeno-associated virus 9; sh, short hairpin RNA; FoxO6, forkhead box O6; AAV9-Ctrl, AAV9-control group.

