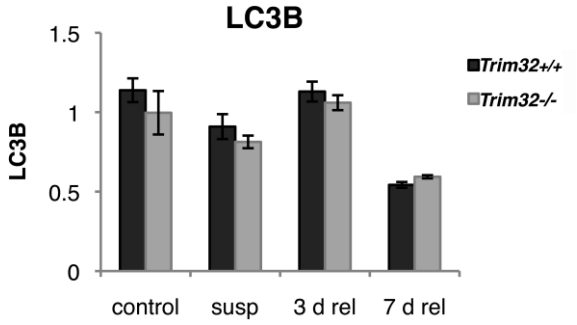
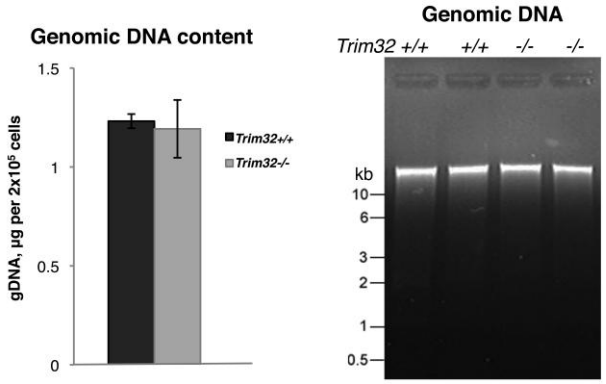


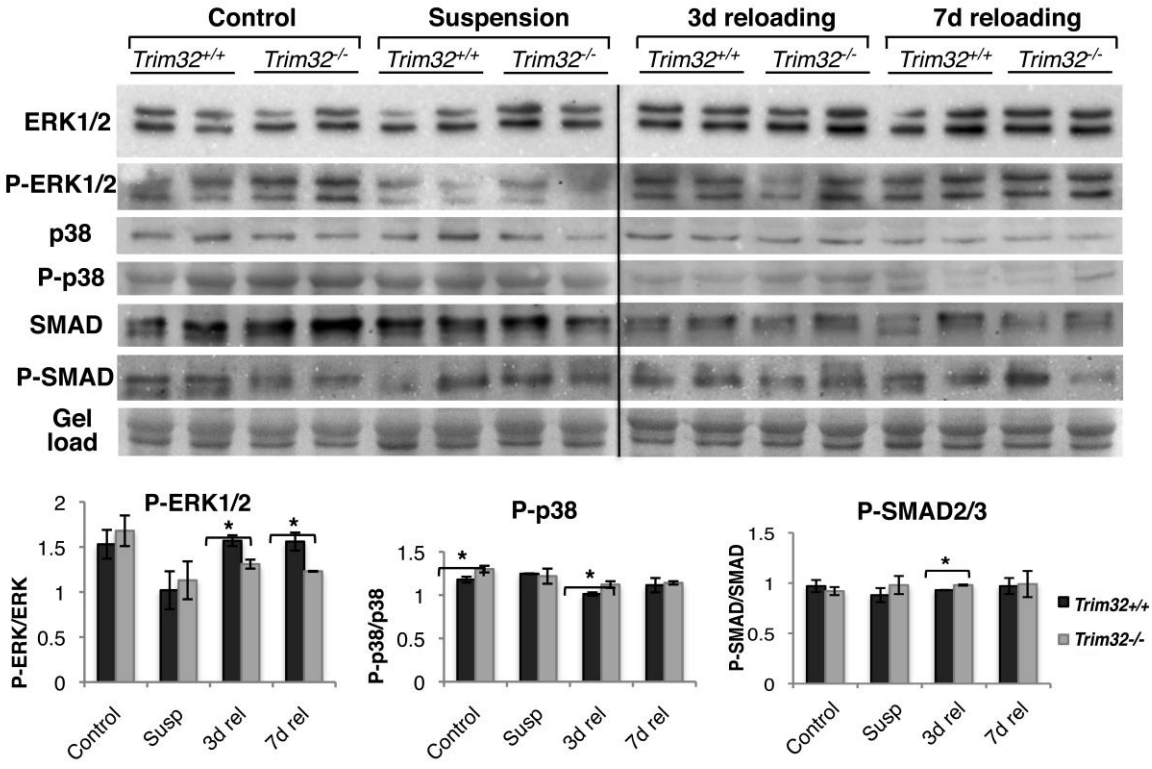

**Supplemental Figure 1**

*Autophagy marker LC3B is not changed in Trim32$^{-/-}$ muscles.*
Densitometry data of western blots of soleus muscle extracts from hind-limb suspension experiment stained with autophagy marker LC3B did not reveal a statistically significant difference between *Trim32$^{-/-}$* and *Trim32$^{+/+}$*. Data are normalized to loading control and shown as mean ± s.e.m.




**Supplemental Figure 2**

*Total amount of genomic DNA is not changed in Trim32$^{-/-}$ myoblasts.* Equal amounts of genomic DNA were isolated from *Trim32$^{-/-}$* and *Trim32$^{+/+}$* myoblasts as revealed by spectrophotometric analysis of gDNA purified from equal amount of *Trim32$^{-/-}$* and *Trim32$^{+/+}$* cells. Data are shown as mean ± s.e.m. Representative agarose gel is shown for quality control of the gDNA preparation.




**Supplemental Figure 3**

*Canonical and non-canonical TGF-β signaling in Trim32$^{-/-}$ and Trim32$^{+/+}$ muscles during unloading/reloading.* Representative western blots of soleus muscle extracts from ambulatory control (control), suspended for 5 d (suspension), and reloaded for 3 or 7 days mice were stained with total ERK1/2, activated ERK1/2-phospho-specific antibodies, total p38, phospho-p38, SMAD2/3, and phospho-SMAD2/3 antibodies. Ponceau S stained blots are shown for loading control. Densitometry data of activated P-ERK1/2, P-p38, and P-SMAD2/3 blots were normalized to total ERK1/2, total p38 or total SMAD2/3, respectively. A modest 11% increase in P-p38, a 5% increase in P-SMAD2/3, and a 17-20% decrease in P-ERK1/2 was observed in *Trim32$^{-/-}$* muscles subjected to reloading, compared to *Trim32$^{+/+}$* muscles. P-p38 was also increased by 10% in *Trim32$^{-/-}$* control muscles, compared to *Trim32$^{+/+}$* littermates. Data are shown as mean ± s.e.m (*p<0.05).