

Additional file 1 for

Heat shock protein A4 ablation leads to skeletal muscle myopathy associated with dysregulated autophagy and induced apoptosis

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Fig. S2. Maintained glucose and lipid metabolism in *Hspa4*-KO mice

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Fig. S4. Myopathy in the paraspinal muscle in 18-month-old *Hspa4*-KO mice.

Supplementary figures

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Fig. S1. HSPA4 protein is ubiquitously expressed in different types of skeletal muscles. Representative immunoblot of HSPA4 protein in skeletal muscles. Dia, Diaphragm; GC, Gastrocnemius; TA, Tibialis anterior.

Fig. S2. Maintained glucose and lipid metabolism in *Hspa4*-KO mice. (A) Serum glucose levels in 2-week-old WT and *Hspa4*-KO mice. (B and C) Northern blot (B) and real time PCR (C) analyses of gluconeogenic enzyme expression, phosphoenolpyruvate carboxykinase 1 (*Pepck*), in 2-month-old liver. (D and E) Northern blotting (D) and real time PCR (E) of hepatic lipid transport related-genes expression, *Sar1b* and *ApoB* in 10-day-old mice. (E) Real time PCR analyses of hepatic *Sar1b* and *ApoB* in 10-day-old mice. (F) Northern blotting analyses of growth hormone-responsive gene expression, solute carrier organic anion transporter (*Slco1b2*), in 10- and 14-day-old liver. (G) *Slco1b2* expression in 14-day-old liver, assessed by real time PCR. Data are expressed as mean \pm SEM, two-tailed unpaired Student's t-test. Numbers within columns indicate mice.

Fig. S3. Increased centrally nucleated myofibers in *Hspa4*-KO soleus muscle. (A) H&E stained-sections of soleus from 3-week-old mice. (B) Increased centrally nucleated fibers in soleus muscle at 2- and 3-week-old *Hspa4*-KO mice.

Fig. S4. Myopathy in the paraspinal muscle in 18-month-old *Hspa4*-KO mice. Note centrally nucleated myofibers (arrows).

Fig. S1

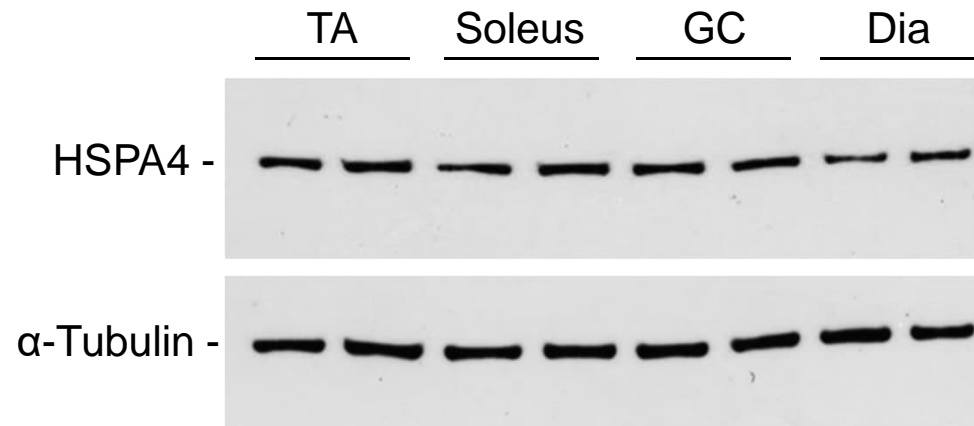


Fig. S2

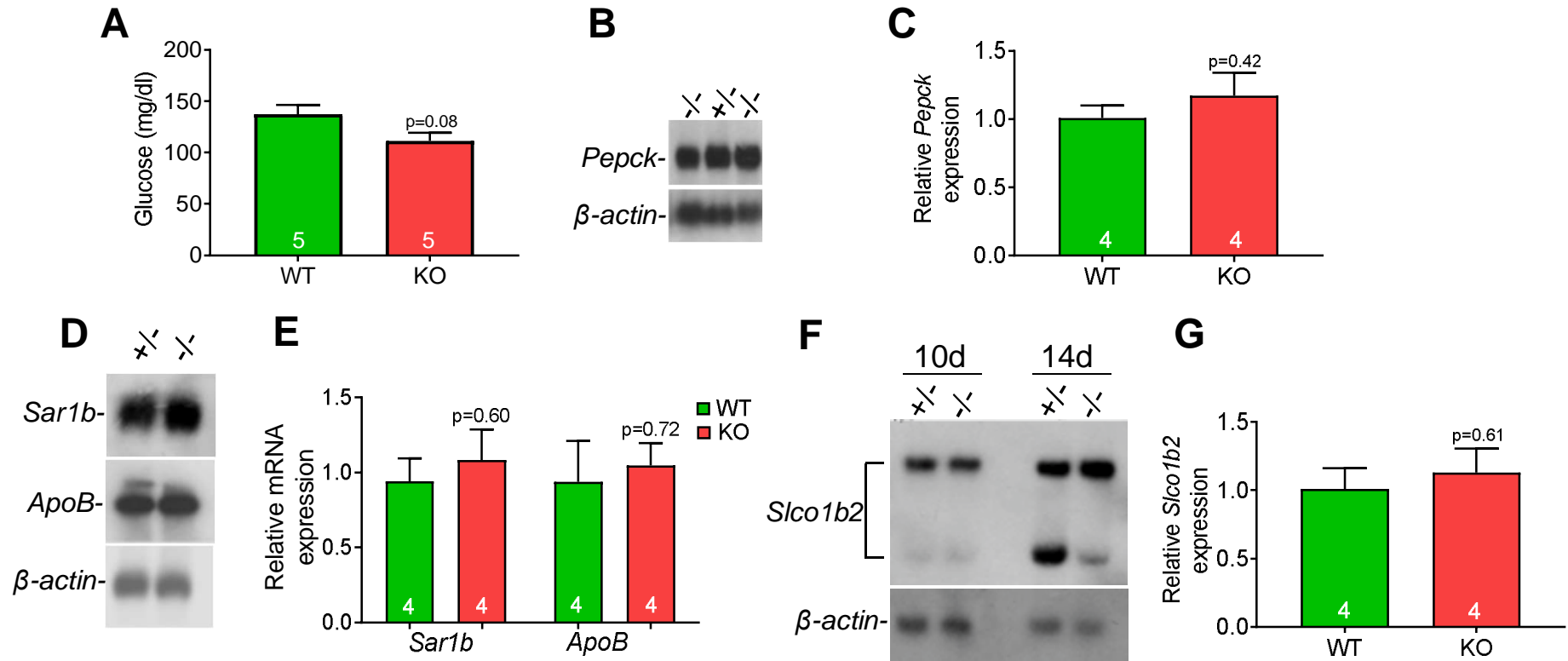


Fig. S3

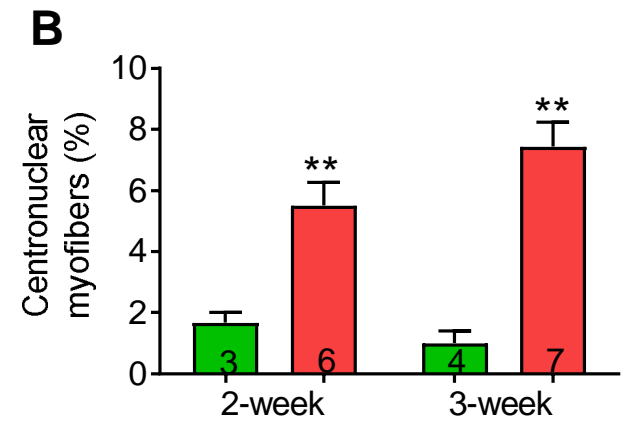
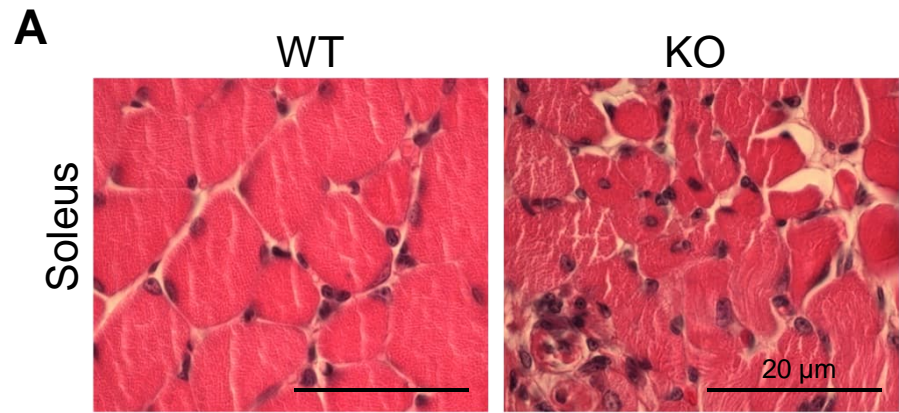


Fig. S4

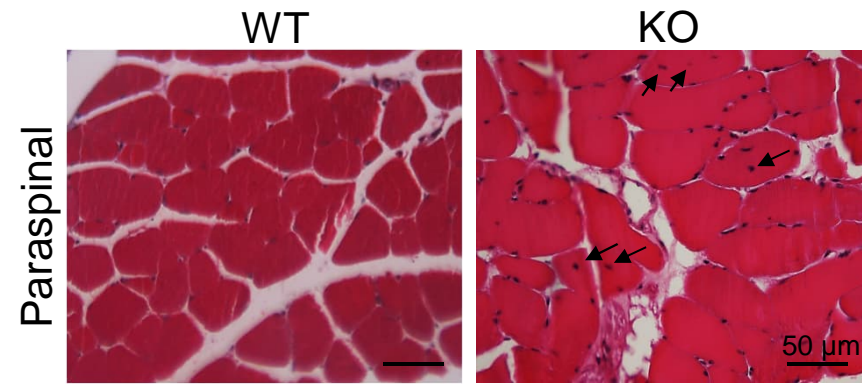


Table S1. List of mouse primers used in this study

Real time PCR	
<i>Hprt1</i>	F: 5`-AGCCCCAAAATGGTTAAGGTTGC-3` R: 5`-TTGCAGATTCAACTTGCCTCAT-3`
<i>Myh8</i>	F: 5`-AACAGAAACGCAATGCTGAGG-3` R: 5`-TCGCCTGTAATTTGTCCACCA-3`
<i>Myh3</i>	F: 5`-AAAAGGCCATCACTGACG-3` R: 5`-CAGCTCTCTGATCCGTGTCTC-3`
<i>MAFbx</i>	F: 5`-CTCTGTACCATGCCGTTCT-3` R: 5`-GGCTGCTGAACAGATTCTCC-3`
<i>MuRF1</i>	F: 5`-ACGAGAAGAAGAGCGAGCTG-3` R: 5`-CTTGGCACTTGAGAGGAAG-3`
<i>Pepck</i>	F: 5`-AGCCTGCTCCAGCTTTGA-3` R: 5`-CCCTAGCCTGTTCTCTGTGC-3`
<i>Sar1b</i>	F: 5`-CAGCACGTCCCAACTACA-3` R: 5`-AAACGTCATGCCAGCAATAGT-3`
<i>ApoB</i>	F: 5`-TGGGATTCCATCTGCCATCTCGAG-3` R: 5`-GTACAGATCCATCACAGGACAATG-3`
<i>Myod1</i>	F: 5`-GCGCTCCAAGTCTCTGATG-3` F: 5`-TCGACACAGCCGCACTCTTC-3`
<i>Myogenin</i>	F: 5`-CCAACCCAGGAGATCATTG-3` R: 5`-GGAAGGCAACAGACATATCCTC-3`
<i>Pax7</i>	F: 5`-GGGATGTTTCCAGCTGGGAAA-3` R: 5`-CACTCGGCTAATCGAACTCA-3`
<i>Cd68</i>	F: 5`-ACCGCTTATAGCCCAAGGAAC-3` R: 5`-CGTGAAGGATGGCAGGAGAGTAA-3`
<i>Il1b</i>	F: 5`-ATGCCACCTTTTGACAGTGATG-3` R: 5`-GCTCTTGTTGATGTGCTGCT-3`
<i>Il6</i>	F: 5`-TCGTGGAAATGAGAAAAGAGTTGTG-3` R: 5`-ATCCAGTTTGGTAGCATCCATCA-3`
<i>F4/80</i>	F: 5`-CCCCAGTGTCTTACAGAGTG-3` R: 5`-GTGCCAGAGTGGATGTCT-3`
Northern blotting	
<i>Pepck</i>	F 5`- GTGGAGGAGATCGACAGGTATC -3` 5`- CTCACACAGAGACACGTTTAC-3`
<i>ApoB</i>	F 5`- CCAACAGAAATGTCCACAGAGA -3` R 5`- AGCCACTGGAGGATGTGAGTAT -3`

Slco1b2

F 5`-TGCGATGGATTCAGGATATT-3'

R 5`-ACCCCTTTTCACAACCTTTC -3'