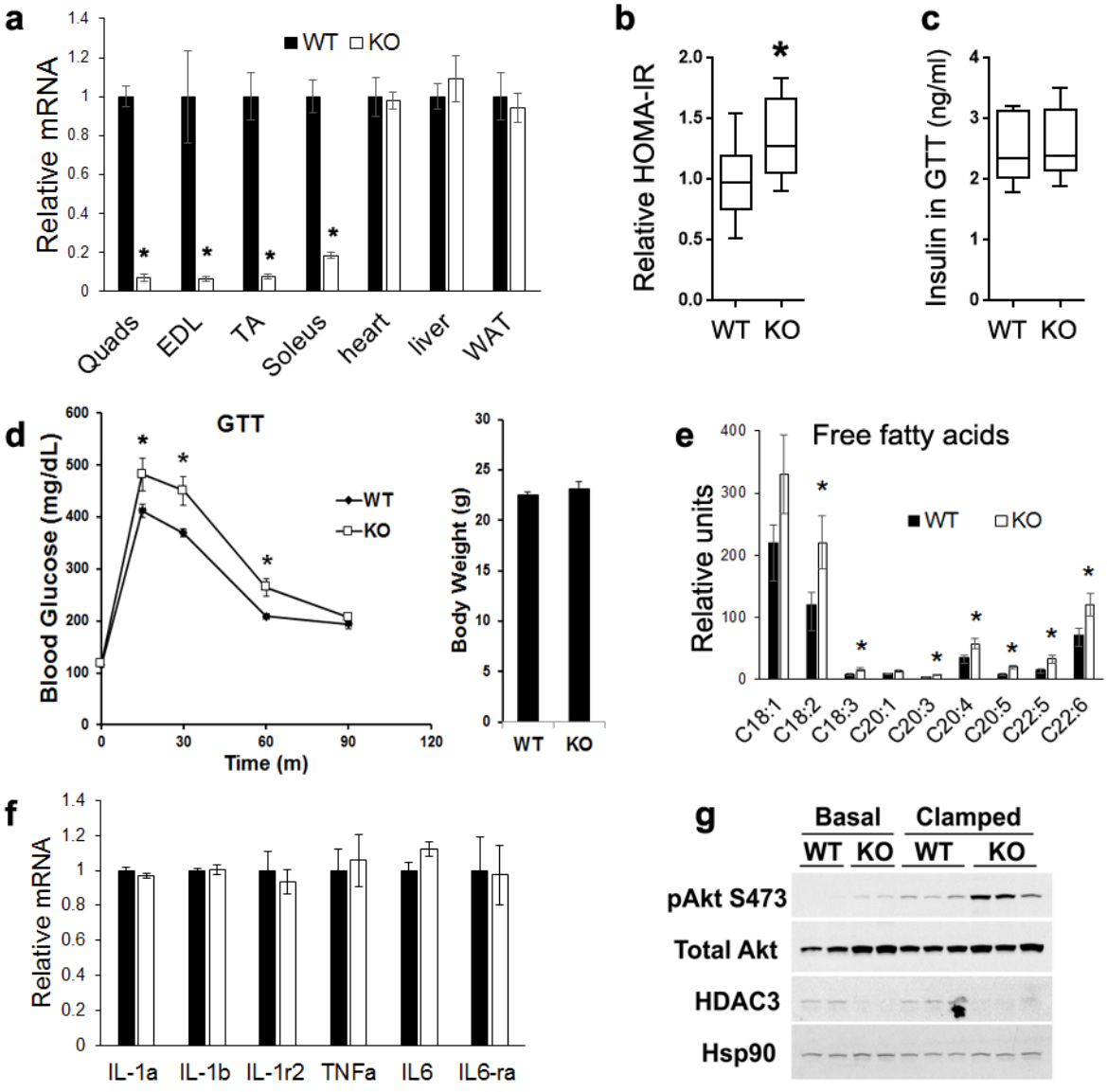
SUPPLEMENTAL FIGURES

Supplementary Figure 1



Supplementary Figure 1. (a) RT-qPCR analysis of HDAC3 at different tissues. Quads, quadriceps; EDL, extensor digitorum longus; TA, tibialis anterior; WAT, white adipose tissue. *n* = 5. **(b)** Relative homeostatic model assessment for insulin resistance (HOMA-IR). Values are basal glucose levels multiplied with basal insulin levels, followed by normalization to the average value of WT mice. *n* = 8. **(c)** Serum insulin levels taken at 30 min during glucose tolerance test (GTT), *n* = 8. **(d)** GTT on 8-weeks old mice on normal chow, *n* = 8. **(e)** Quantification of free fatty acids in quadriceps muscle by mass spectrometry, *n* = 5. **(f)** RT-qPCR analysis of inflammatory genes in quadriceps, *n* = 5. **(g)** Western blot analysis of molecular insulin signaling in muscles harvested immediately after the insulin clamp assay. Data were presented as the mean ± S.E.M. * $P < 0.05$ between genotypes by t-test.

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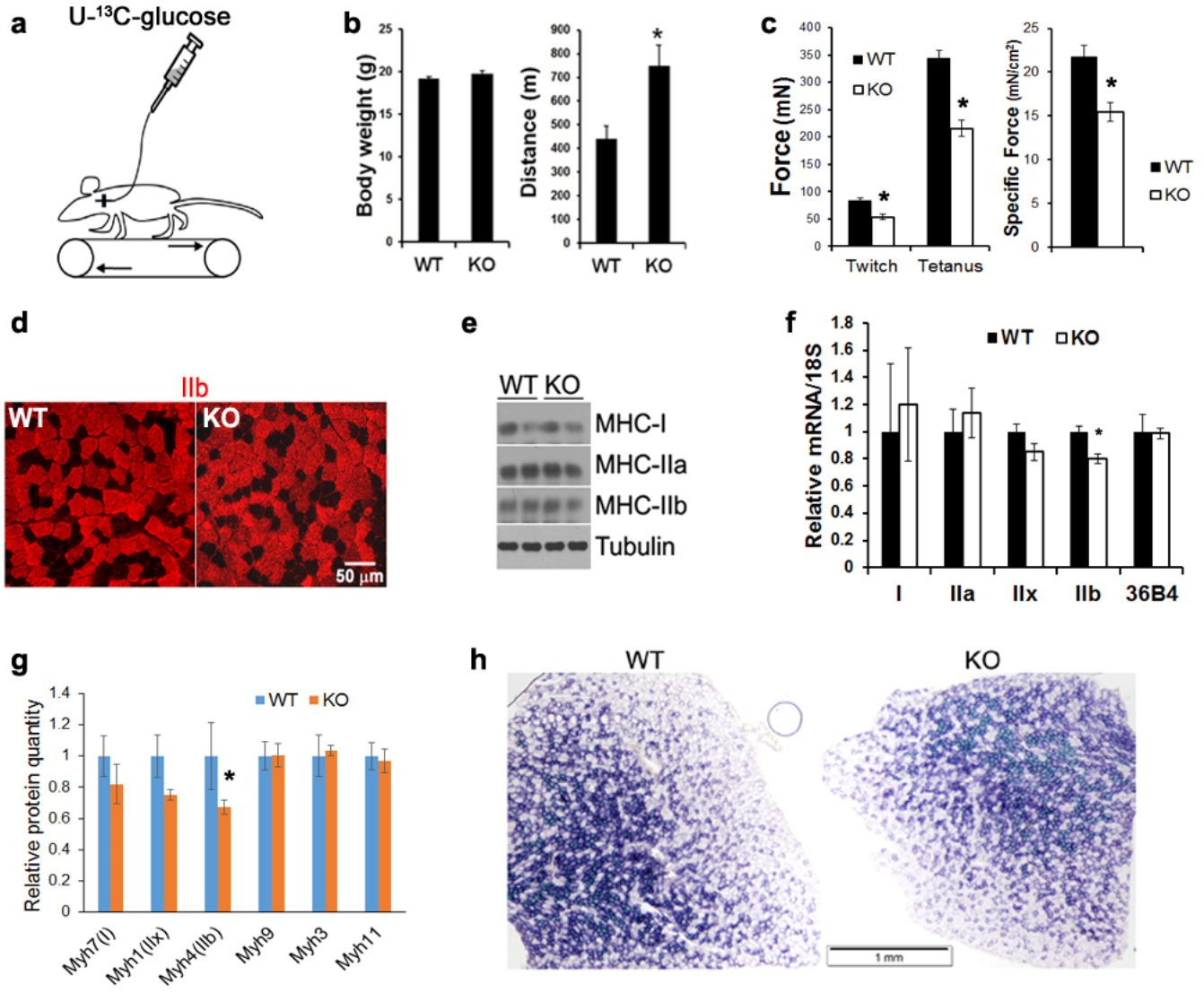
Supplementary Figure 2



Supplementary Figure 2. (a) Diagram showing the set-up of the experiment. Mice were infused with ^{13}C-glucose through jugular vein while running on treadmill. **(b)** Body weight and distance run on treadmill at the time of exhaustion (50 electric shocks received) for female mice at 8-weeks old, *n* = 10. **(c)** Muscle force measurement during *ex vivo* contraction studies with extensor digitorum longus (EDL) muscles, *n* = 8. **(d)** Immunofluorescence staining of cross sections of TA muscles with antibodies for MHC IIb. **(e)** Western blot analysis with myosin heavy chain (MHC) isoform-specific antibodies in gastrocnemius muscles. **(f)** RT-qPCR analysis of the indicated MHC isoforms in gastrocnemius at 8-months old, *n* = 5. **(g)** Nano-LC-MS-based TMT proteomic analysis of myosin isoforms in gastrocnemius 6 months old, *n* = 3. **(h)** Succinate dehydrogenase (SDH) staining of TA muscles. Data were presented as the mean ± S.E.M. * $P < 0.05$ between genotypes by t-test.

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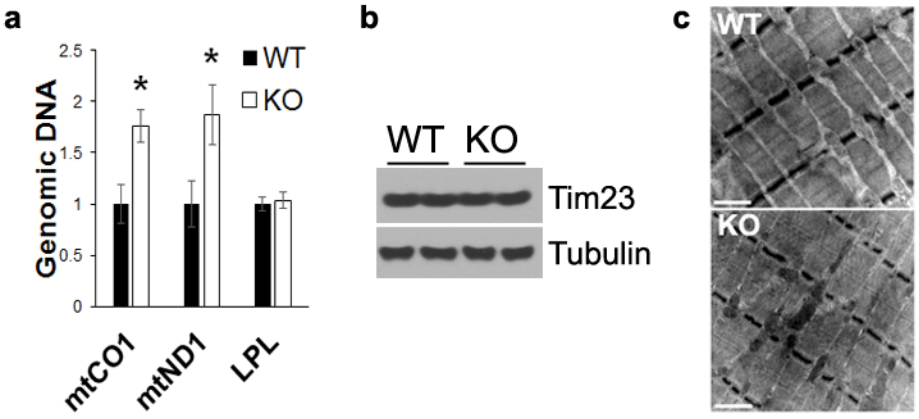
Supplementary Figure 3



Supplementary Figure 3. (a) Mitochondrial DNA quantification by qPCR in whole genome extraction of EDL muscles. Mitochondrial genes (mtCO1 and mtND1) were normalized to a nuclear gene Ndufv1. Another nuclear gene Lpl serves as an independent control, n = 5. **(b)** Western blot analysis of quadriceps with antibodies for mitochondrial marker protein Tim23. Tim23, translocase of the inner membrane 23. **(c)** Electron microscopy of EDL muscle (scale bar: 1 um). Data were presented as the mean ± S.E.M. * $P < 0.05$ between genotypes by t-test.

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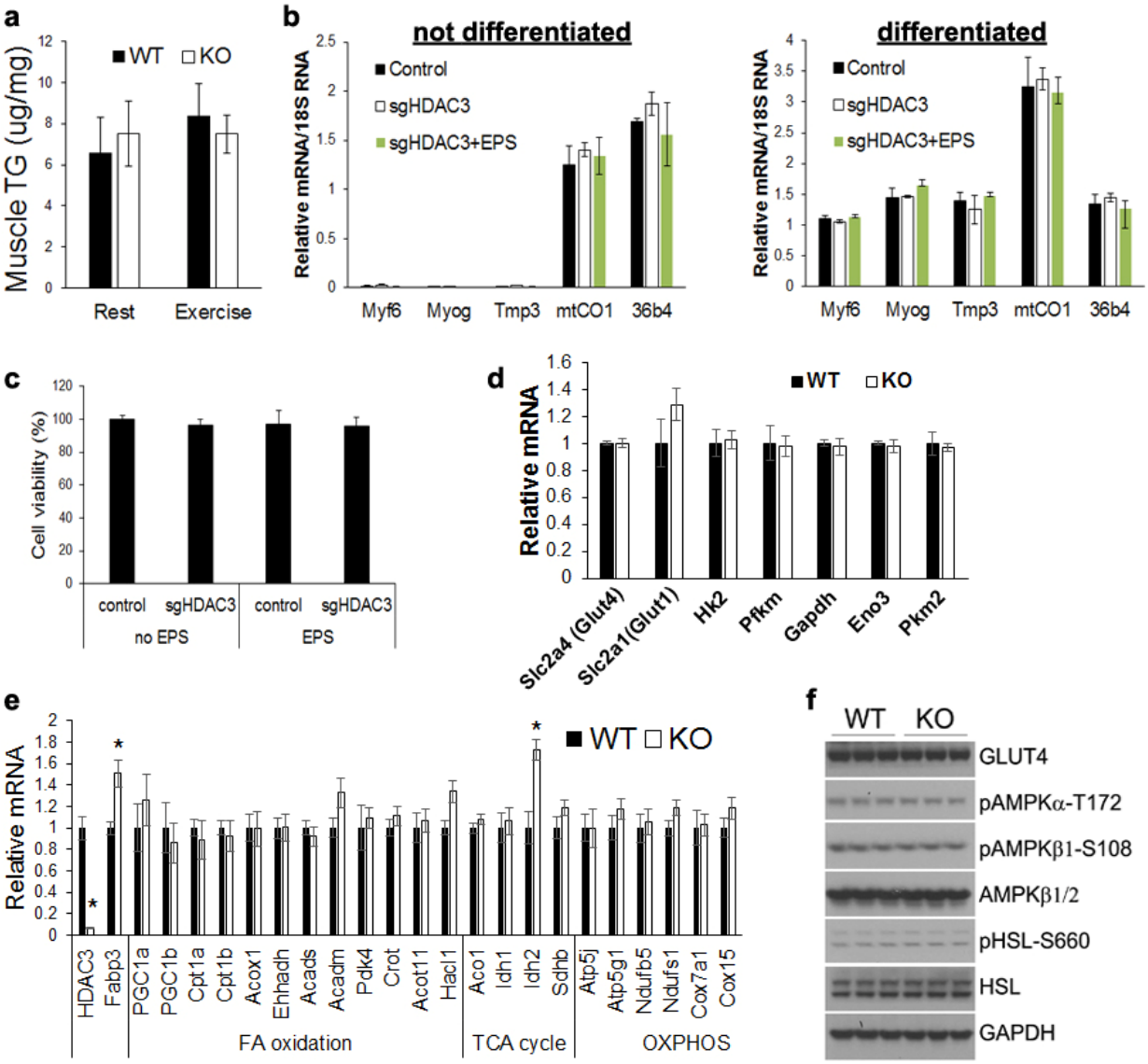
Supplementary Figure 4



Supplementary Figure 4. (a) Muscle triglycerides (TG) measurement, *n* = 5. **(b)** RT-qPCR analysis of differentiation marker genes in C2C12 myotubes treated with adenovirus and/or electric pulse stimulation (EPS). 36b4 serves as a 2nd housekeeping gene control in addition to 18s RNA. *n* = 5. **(c)** MTT cell viability assay of C2C12 myotubes treated with adenovirus and/or electric pulse stimulation (EPS), *n* = 5. **(d)** RT-qPCR analysis of genes involved in glucose transport and metabolism in tibialis anterior (TA) muscles, *n* = 5. **(e)** RT-qPCR analysis of major fatty acid oxidation genes and OXPHOS genes in quadriceps muscles, *n* = 5. **(f)** Western blot analysis of GLUT4 transporter, AMP-activated protein kinase (AMPK), and AMPK substrate hormone sensitive lipase (HSL). Data were presented as the mean ± S.E.M. * *P* < 0.05 between genotypes by t-test.

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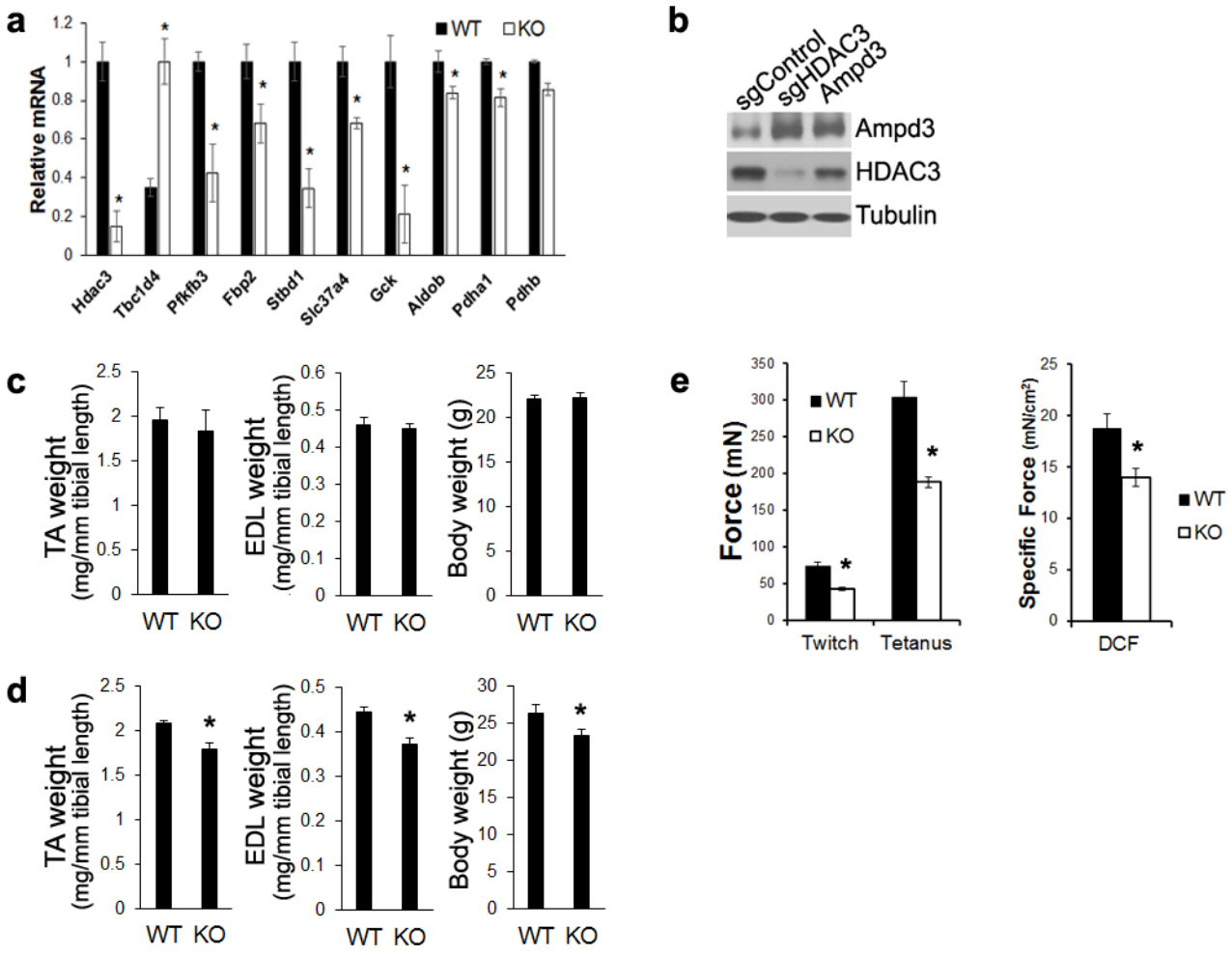
Supplementary Figure 5



Supplementary Figure 5. (a) RT-qPCR analysis of genes involved in carbohydrate metabolism in tibialis anterior (TA) muscles, *n* = 5. **(b)** Western blot analysis of differentiated C2C12 myotubes treated with adenovirus for knockdown of HDAC3 or overexpression of Ampd3. **(c)** Muscle weight and body weight in 6-month old female mice. TA, tibialis anterior; EDL, extensor digitorum longus (EDL); *n* = 8. **(d)** Muscle weight and body weight in 10-month old female mice, *n* = 8. **(e)** Muscle force measurement during *ex vivo* contraction studies with extensor digitorum longus (EDL) muscles in the presence of deoxycoformycin (DCF), *n* = 8. This experiment was performed using the same cohort of mice as shown in supplementary figure 2C. Data were presented as the mean ± S.E.M. * $P < 0.05$ between genotypes by t-test.

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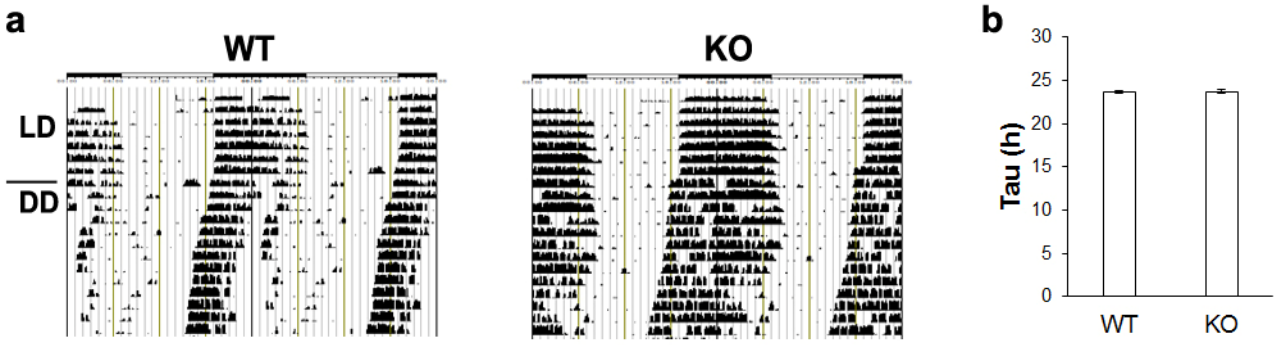
Supplementary Figure 6



Supplementary Figure 6. (a) Actogram of free wheel-running activity of mice housed in normal 12-light/12-dark conditions (LD) and constant darkness (DD). **(b)** Internal period (Tau), *n* = 10. Data were presented as the mean ± S.E.M.

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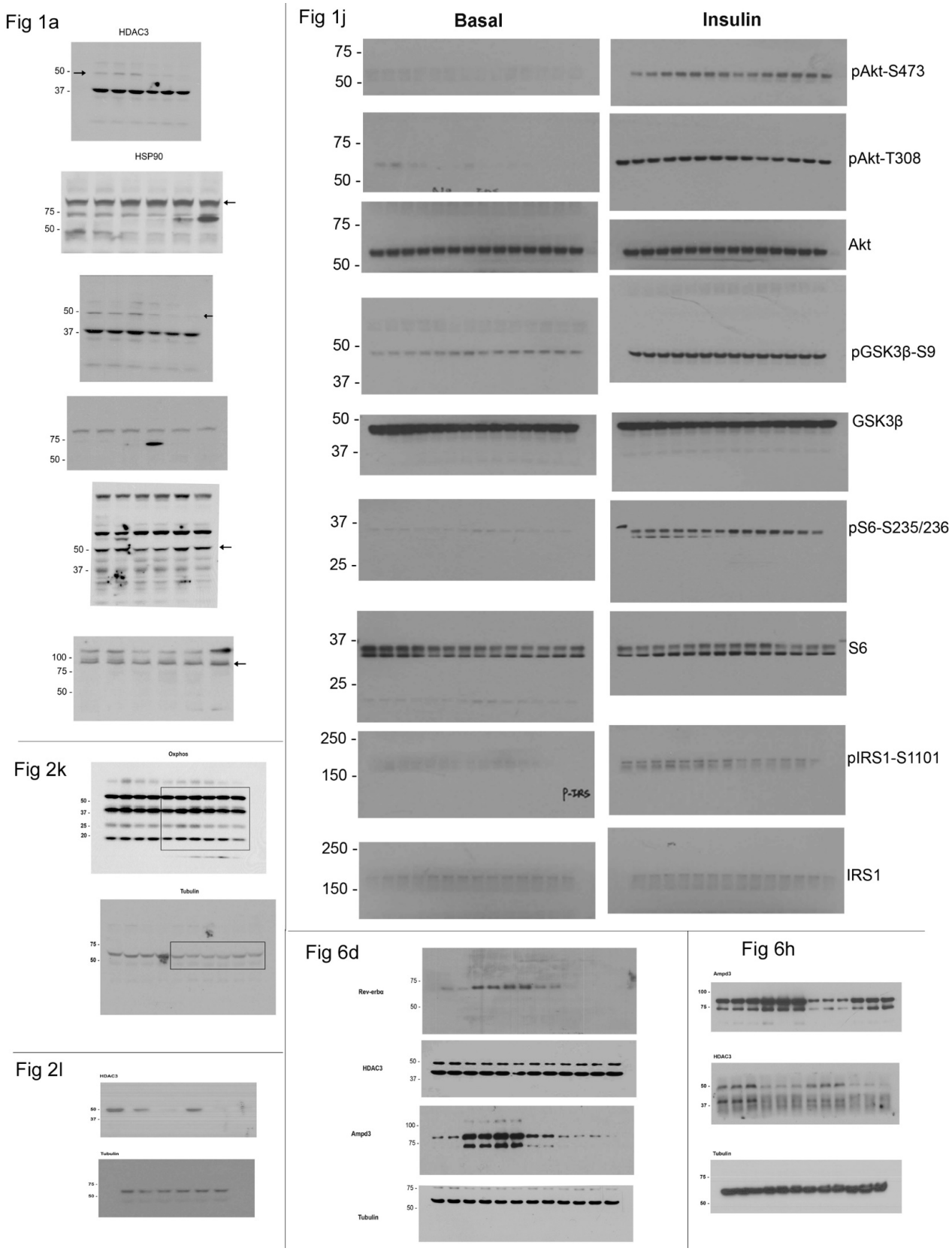
Supplementary Figure 7. Raw gel images for western blot.



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SUPPLEMENTAL TABLES

The following supplemental tables were provided as Supplementary Datasets in one single .xls file.

Supplementary Table 1. Raw data of metabolomics study with ^{13}C-glucose tracing.

Supplementary Table 2. A list of differentially-expressed genes from RNA-seq analysis.

Supplementary Table 3. A list of differentially-expressed proteins from proteomics analysis.

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