High fat diet rescues disturbances to metabolic homeostasis and survival in the *Id2* null mouse in a sex-specific manner.

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SUPPLEMENARY INFORMATION

Supplemental Materials and Methods

Fat mass analysis. *In vivo* X-ray Micro-Computed Tomography (MicroCT) was used to quantify percent body fat Image acquisitions were obtained by using an Albira PET/SPECT/CT image station (Bruker Molecular Imaging, Billerica, MA) [1,2]. WT and *Id2-/-* mice were anesthetized by isofluorane (2.5% flow rate) and maintained at 2.5% setup during imaging. The X-ray source was set to a field of view of 115 mm at current of 200 μA and voltage of 45 kVp. The acquisitions were performed using 600 projections, and reconstruction was completed at a 125 μm voxel size.

PMOD analysis software (PMOD Technologies LTD, Zurich, Switzerland) was used for image analysis. Images were segmented in PMOD following the protocol as described [1]. Total body volume was calculated in cm^3 by segmentation with a range of -300 to +3500 HU. Total body volume was then segmented with a range of -300 to -50 HU to extract only the fat tissue within the mouse. A lower abdominal fat volume was defined as the area after the proximal end of the L1 lumbar vertebrae, and volumes of interest (VOI) of mice were delineated [3]. VolView v3.2 (Kitware, Clifton Park, NY) was used to generate 3D visual displays of segmented images [1].

Analysis of serum and liver lipid and endocrine panels. At the age of 32-34 wk and 22 wk of the feeding experiment, animals were fasted for 16 hr and blood was collected by an intracardiac puncture, drawn at ZT4. Blood was clotted in microtainer tubes (BD, Franklin Lakes, NJ, USA) for 45 min and serum extracted (7500g at 4 ºC for 5 min). After separation, serum was stored at -80 ºC. At the time of sacrifice, liver tissue was harvested and immediately frozen with liquid nitrogen and stored at -80 ºC. Lipid and endocrine panels were generated by University of California at Davis Mouse Metabolic Phenotyping Center (MMPC; Davis, CA). Briefly, serum triglyceride (TG), total cholesterol, HDL cholesterol and LDL cholesterol were measured by spectrophotometry with standard reagents from Fisher Diagnostics (Middletown, VA) (TR22203, TR13303, TR39601, TR53202). Insulin and leptin were measured by electrochemiluminescence (Meso Scale Discovery Rockville, MD). Total adiponectin and Glucagon were measured by Radioimmunoassay from Millipore (Billerica, MA). Liver TG and total cholesterol content was determined by the Folch method [4]. The lipid was reconstituted in isopropyl alcohol and assayed by spectrophotometry with enzymatic reagents from Fisher Diagnostics.

Supplemental References

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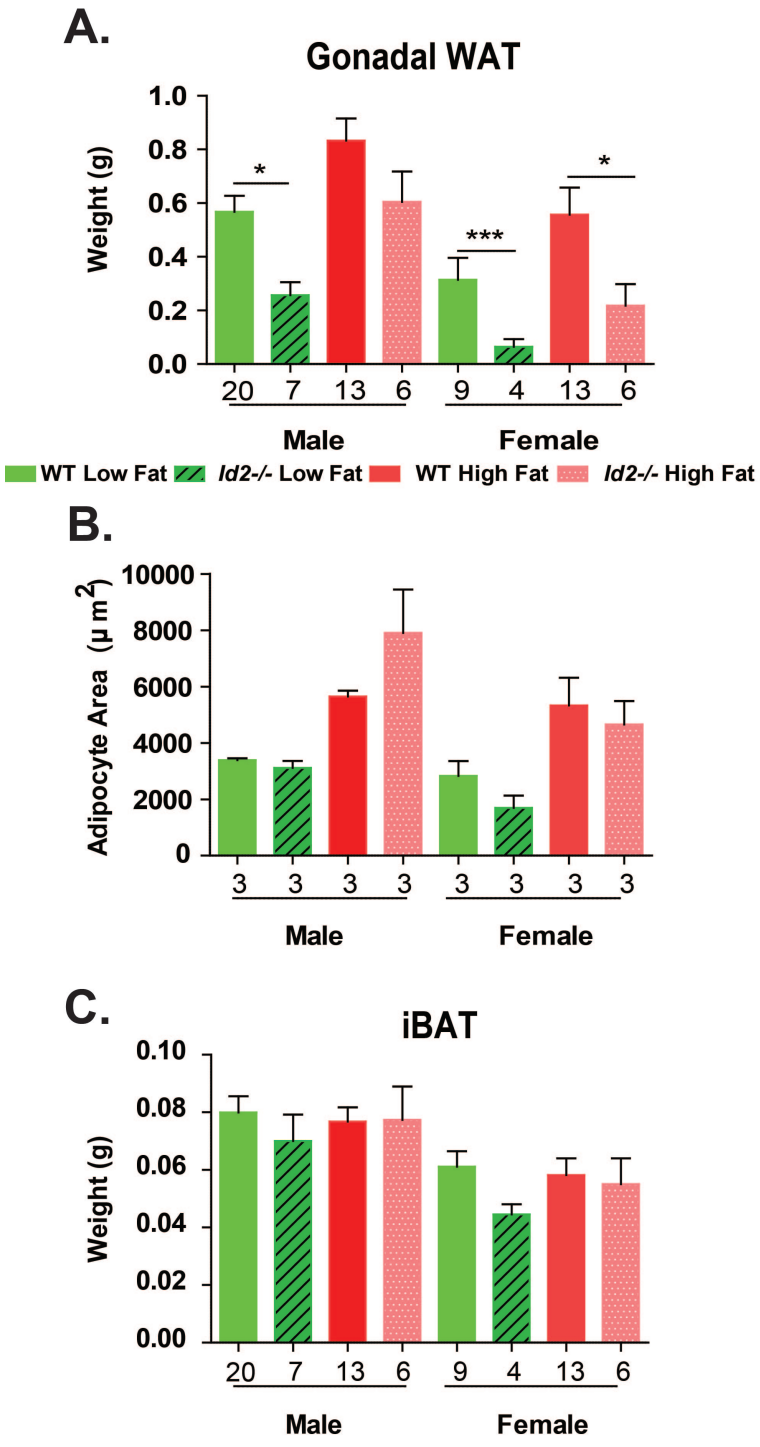
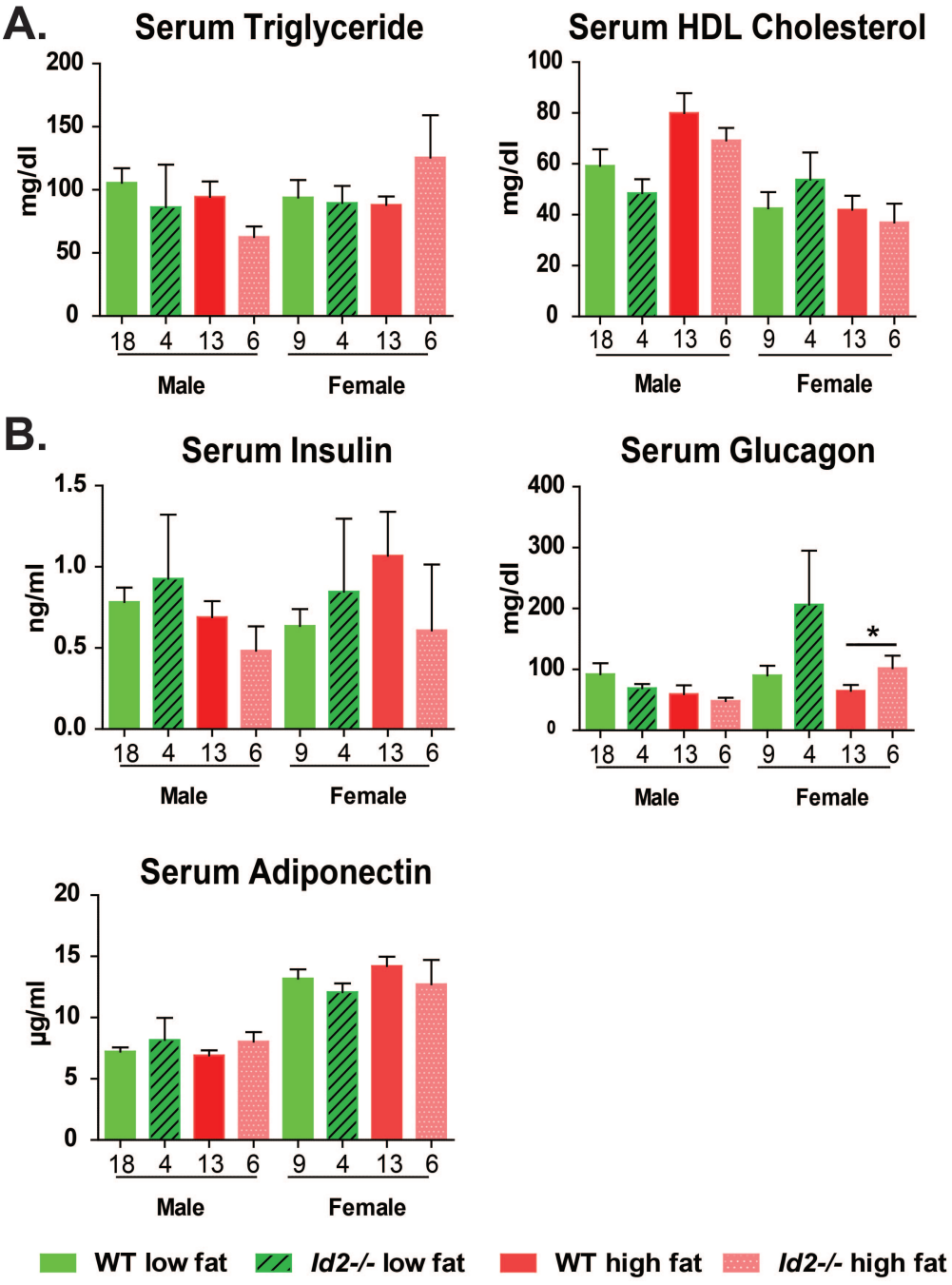


Supplementary Fig. 1. Adipose tissue data. (A) Gonadal white adipose tissue (WAT) mass (g) from WT and *Id2-/-* mice under LFD or HFD. (**B)** Cell area of gonadal WAT in WT and *Id2-/-* mice under LFD or HFD. **C**) interscapular brown adipose tissue (iBAT) mass (g) from WT and *Id2-/-* mice under LFD or HFD. Values are mean ± S.E.M. Two factor-ANOVAs were performed followed by Tukey's post-hoc tests, **p*<0.05, ****p*<0.001.



Supplementary Fig. 2. Serum and liver lipid/endocrine parameters. (A) Serum triglyceride and HDL cholesterol from WT and *Id2-/-* mice under LFD or HFD. (**B)** Serum insulin, glucagon and adiponectin from WT and *Id2-/-* mice under LFD or HFD. Values are mean ± S.E.M. Two factor-ANOVAs were performed followed by Tukey's post-hoc tests, *p<0.05.