

Supplemental material

Supplementary Figure I: Lipid analysis of Hearts by TLC. Lipids from 30 mg hearts of Wild type (WT) mice and MHC-Plin5 mice were extracted and were analyzed by TLC. A mixture of TAG, sn-1.2/1.3DG, MG was used as standard (S). Lipids were visualized with iodine vapors. Age of fasted male animals at the time of analysis was 16 weeks.

Supplementary Figure II: Plin5 is present at the lipid surface in cardiac muscles and highly increased in the fat cake fraction from 4 month-old fed MHC5-Plin5 mice .

a. Representative Immunofluorescence staining was performed on heart cryosections with indicated antibodies. **b.** Supernatant and fat cake fractions were obtained from cardiac muscles from Wild type (WT) and MHC-Plin5 mice as described in the material and methods, Respectively 40µg of supernatant samples and 20µg of fat cake fraction were loaded on the gel. Westernblots were performed as described in material and methods.

Supplementary Figure III. Plin4 presence is increased in MHC-Plin5 fat cake fraction. Supernatant and fat cakes were prepared as described in the material and methods section. Respectively 40µg of supernatant samples and 20µg of fat cake fraction were loaded on the gel. Westernblots were performed as described in material and methods and antibodies against Plin4 were used.

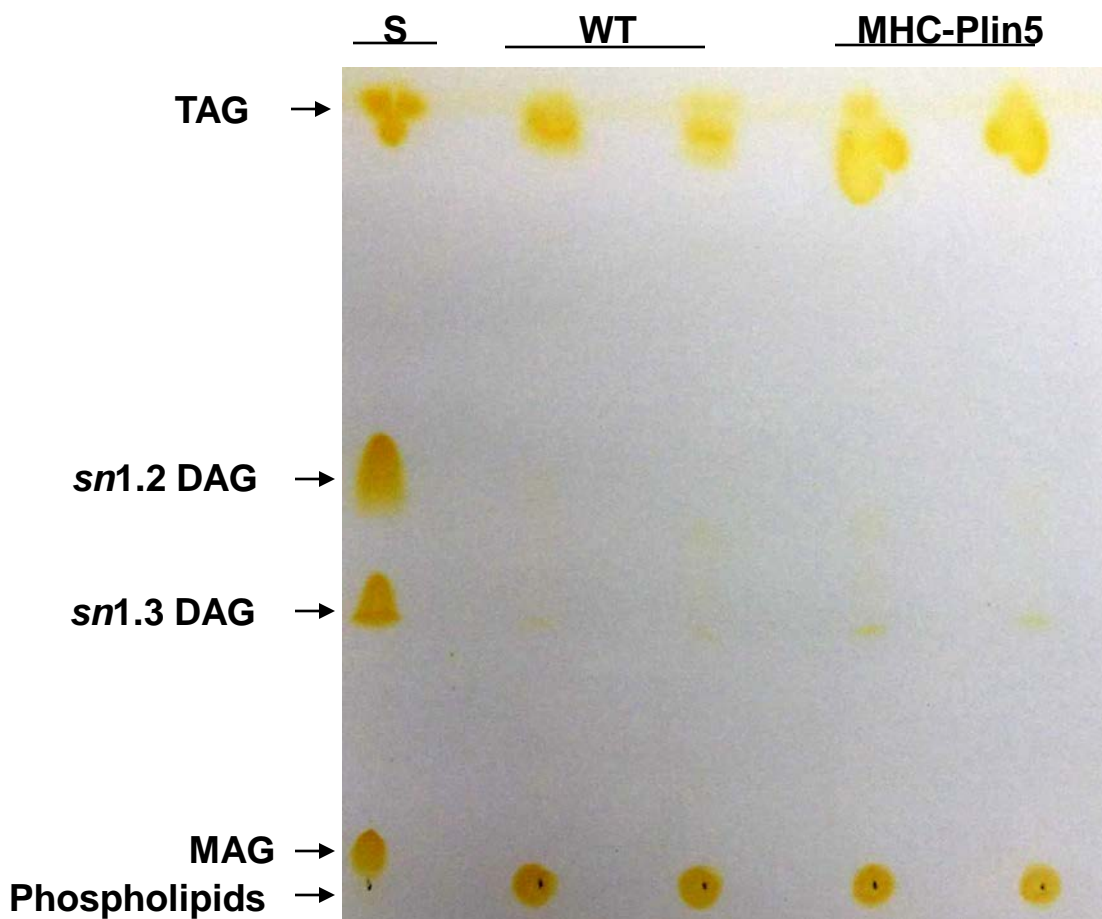
Supplementary Figure IV: mRNA and protein expressions of ATGL and CGI-58, key proteins in LD hydrolysis between MHC-Plin5 and WT left ventricles. **a.** mRNA expression levels in fed 3-month old WT and MHC-Plin5 hearts for ATGL and CGI-58 were determined by RT-qPCR analysis. MHC Plin5 hearts. means are errors bars \pm SEM; n = 9 for each group. **b.** Left ventricles were excised and equal mg equivalent of wet tissue were loaded in each lane. Experiments were performed twice. A commercially available antibody against mouse ATGL was used. **c and d.** Supernatant and fat cakes were prepared as described in the material and methods section. Respectively 80µg of supernatant samples and 40µg of fat cake fraction were loaded on the gel. Westernblots were performed as described in material and methods and antibodies against ATGL (**c**) or against CGI-58 (**d**) were used.

Supplementary Figure V: Morphological abnormalities observed in selected Plin5 MHC mitochondria. Representative electron micrographs depict the histological appearance of left ventricles from fasted WT (A) and MHC-Plin5 (B-D) mice at 4 month-old old (respective magnification x 21000 (A-C), and x 6500 (D) bar equals 100 nm (A-C) and 500 nm (D)). Signs of cristae disarray and intra mitochondrial vacuoles are present in MHC-Plin5 heart images and are signaled by black arrows.

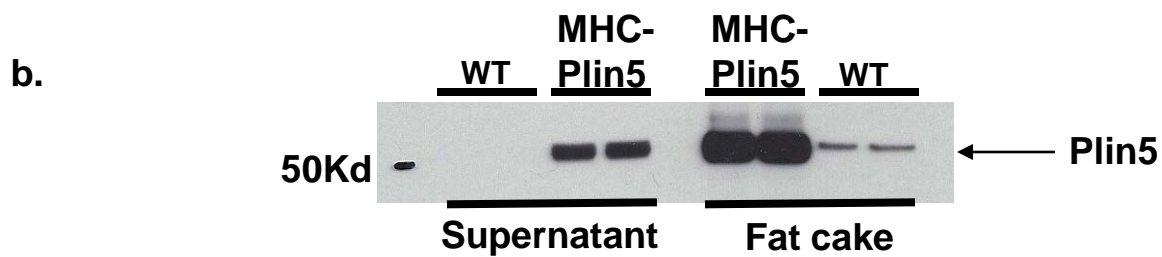
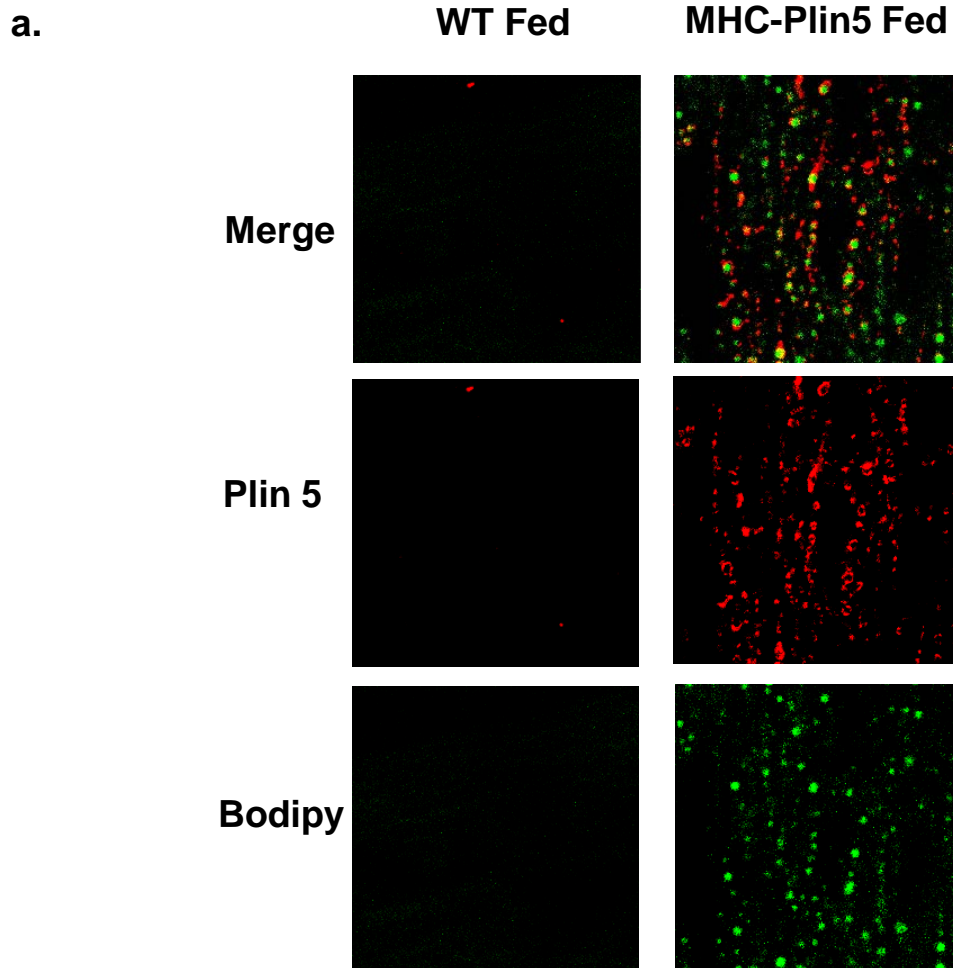
Supplementary Figure VI: During isolation of mitochondria subpopulations, supernatant retains more mitochondria in MHC-Plin5 samples, Representative westernblot of proteins contained increased VDAC, a marker for mitochondria in MHC-Plin5 supernatant left over after during mitochondria isolation compare to WT samples.

Supplementary Figure I

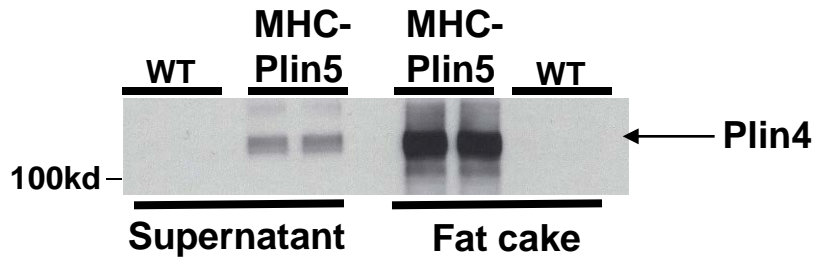
Heart



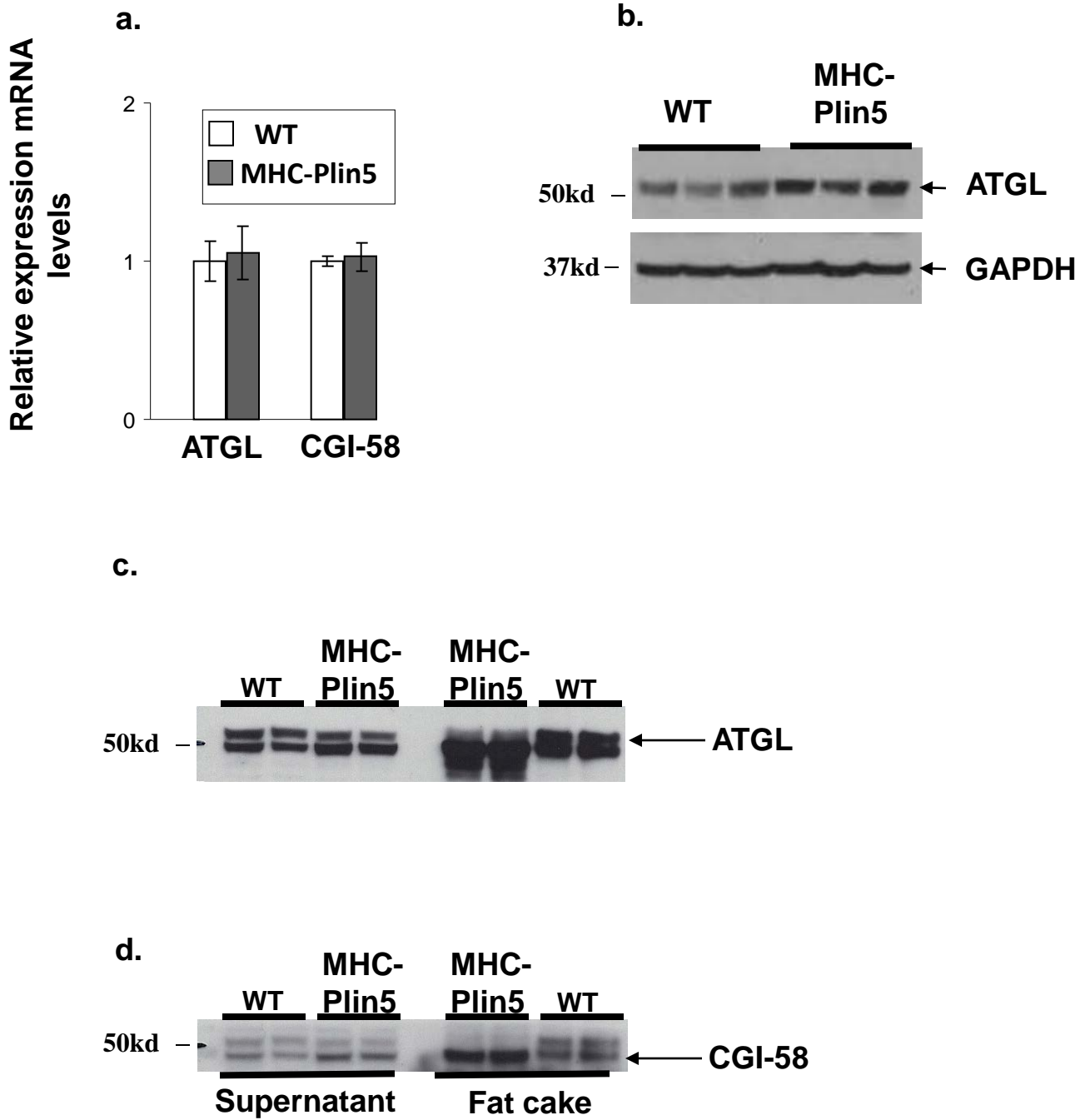
Supplementary Figure II



Supplementary Figure III

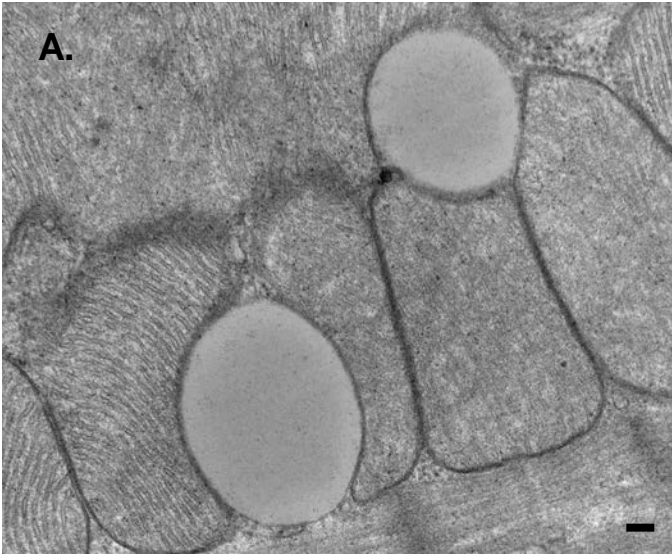


Supplementary Figure IV

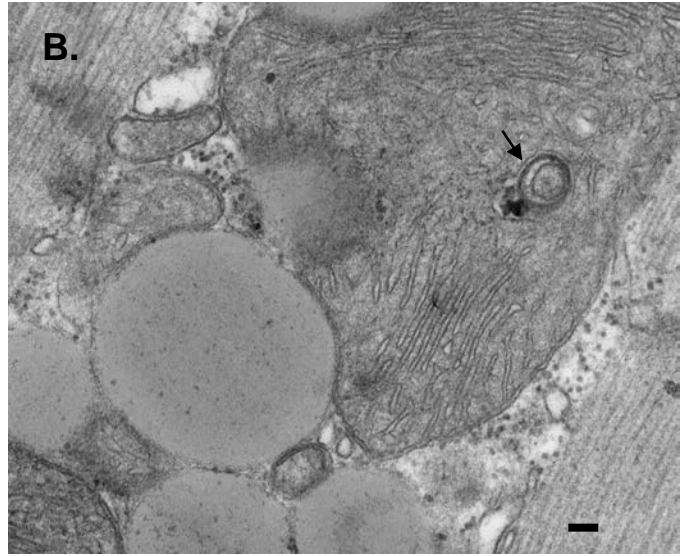


Supplementary Figure V

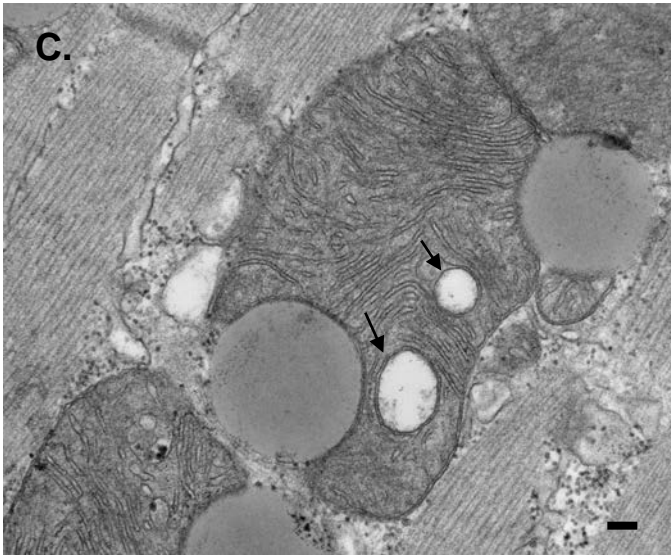
WT



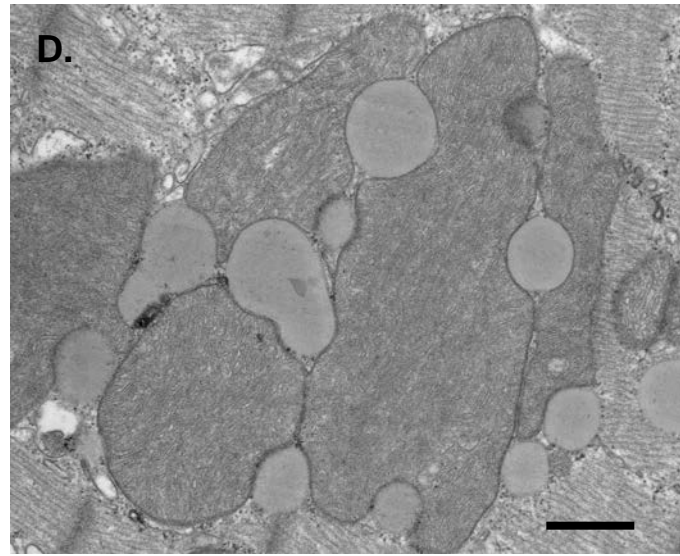
MHC-Plin5



MHC-Plin5



MHC-Plin5



Supplementary Figure VI

