

Supplemental Data

Mitochondrial calcium uniporter in *Drosophila* transfers calcium between the endoplasmic reticulum and mitochondria in oxidative stress-induced cell death

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Supplementary Figure Legends

FIGURE S1. Comparison of MCU sequences among human, mouse and *Drosophila*. *A*, amino acid sequence alignments of *Drosophila* MCU (dMCU), human MCU (hMCU) and mouse MCU (mMCU).

FIGURE S2. Mitochondrial localization of MTRP. Subcellular localization of MTRP in larval muscle *A*, MTRP is green. Hoechst (blue) and ATP5a (red) were used to mark DNA and mitochondria, respectively. Genotype: *Mef-Gal4/UAS-MTRP*. Scale bar: 10 μ m.

FIGURE S3. Cytosolic calcium changes by caffeine in *MCU⁵²* mutant. *A*, averaged traces (left) and the quantitative analysis (right) of $[Ca^{2+}]_i$ signals after treatment of 10 mM caffeine from wild-type and *MCU⁵²* mutant (n=3~4). Genotypes: *Mef>* (*Mef-Gal4/UAS-GCaMP3.T*) and *MCU⁵²,Mef>* (*Mef-Gal4,MCU⁵²/UAS-GCaMP3.T,MCU⁵²*).

FIGURE S4. Oxidative stress-induced apoptosis in *MCU⁵²* mutant. *A*, ROS level change was detected by DHE staining in thoraces of adult fly feeding on normal or 1% H₂O₂ containing food for 72 hours. DHE is red. Hoechst (blue) used to mark DNA. Scale bars: 20 μ m. *B*, the proportion of apoptotic nuclei expressing the GFP reporter under control of *hid5*'*F-WT* enhancer in wild-type and *MCU⁵²* mutant flies after treatment of 2% H₂O₂ for 3 hours (n=5~7). *C*, the proportion of apoptotic nuclei stained with cleaved caspase-3 in wild-type and *MCU⁵²* mutant flies after treatment of 2% H₂O₂ for 3 hours (n=5). ** and *** denote p<0.01 and <0.001, respectively.

FIGURE S5. Calcium changes by t-BuOOH. *A*, averaged traces (left) and quantitative analyses (right) of $[Ca^{2+}]_{mito}$ signals after 1, 10, 35 and 100 mM t-BuOOH treatment (n=4~6). Genotype: *Mef-Gal4/UAS-MTRP*. *B*, averaged traces (left) and quantitative analyses (right) of $[Ca^{2+}]_i$ signals after 35 mM t-BuOOH treatment in control (*Mef>*) and *MCU⁵²* flies (n=5~7).

Supplementary Figures

Figure S1

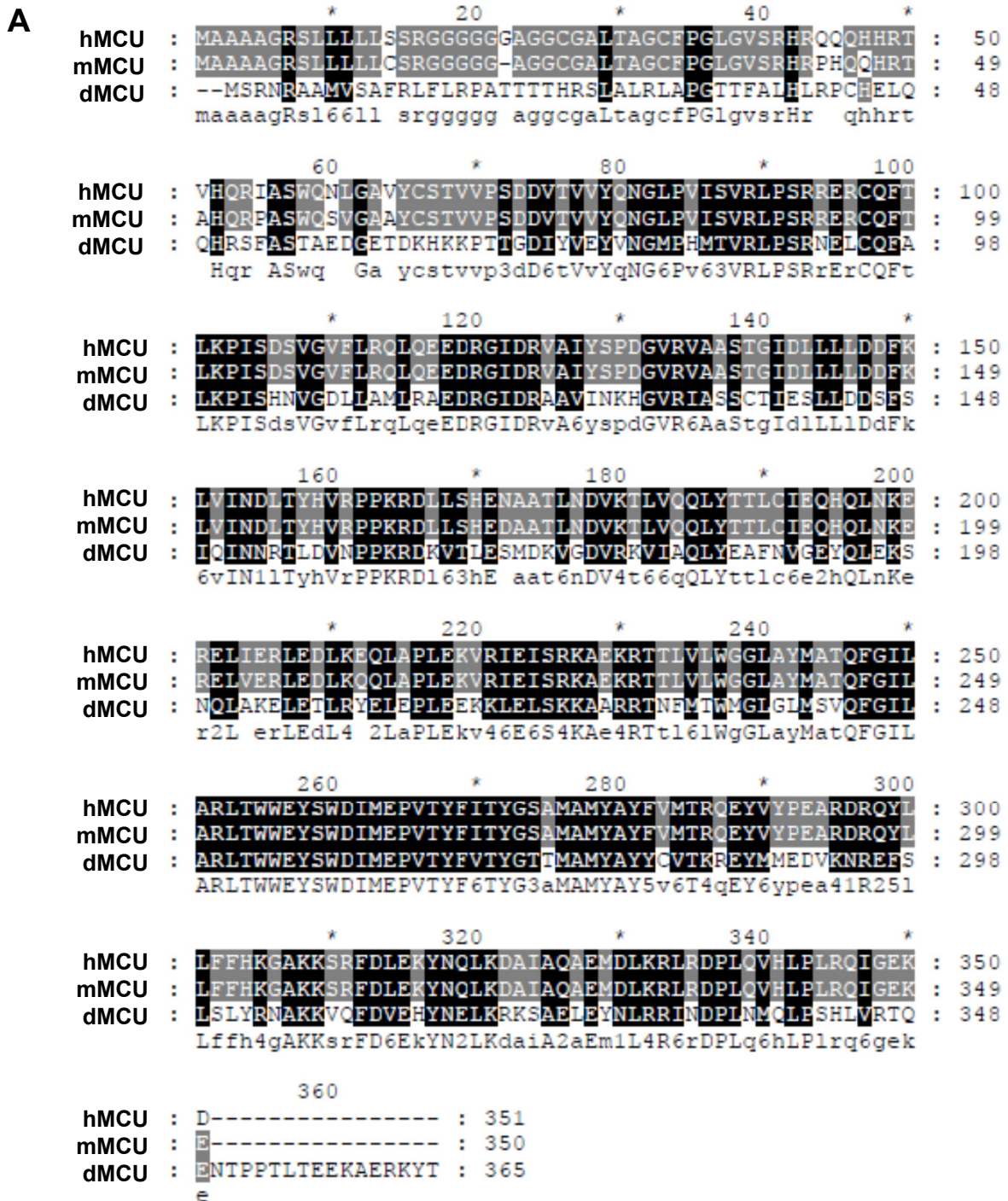


Figure S2

A

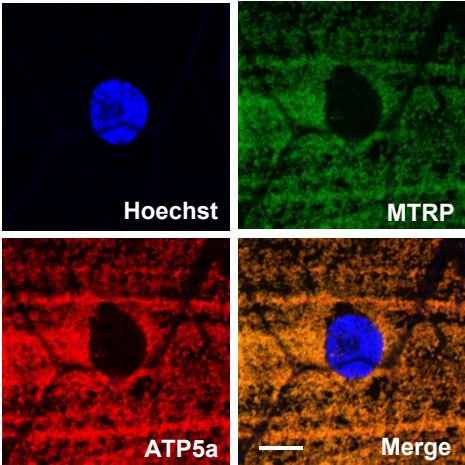


Figure S3

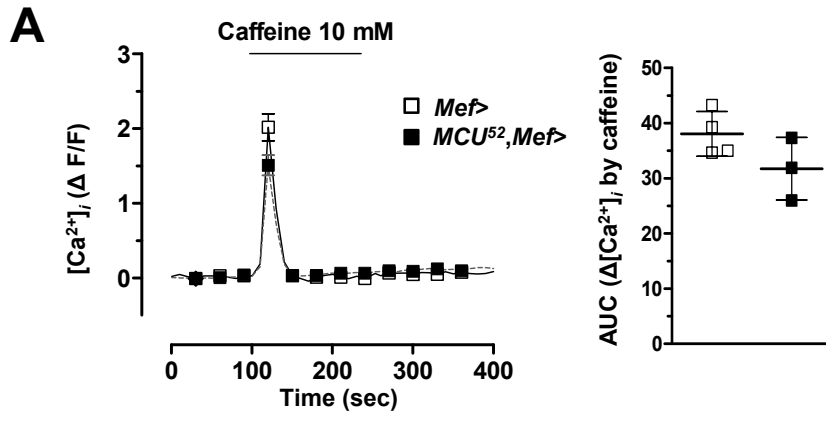
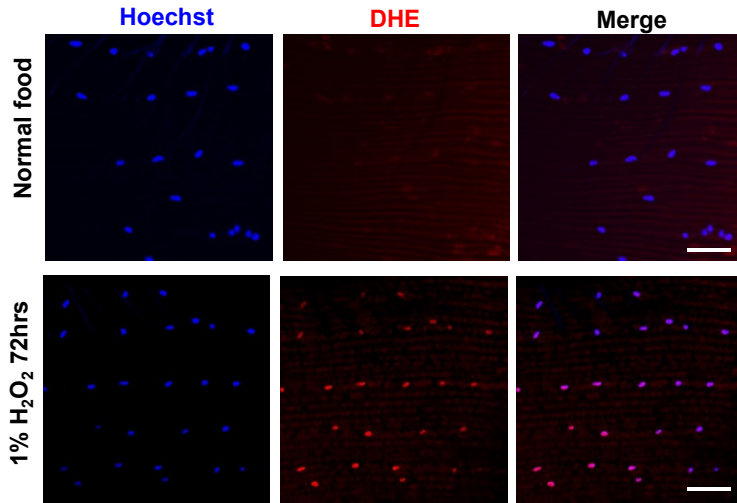
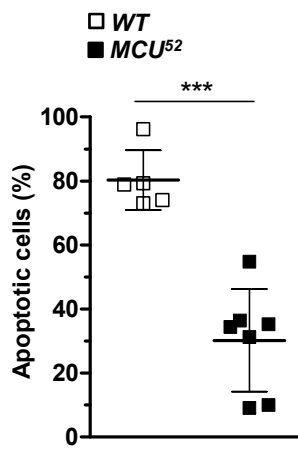


Figure S4

A



B



C

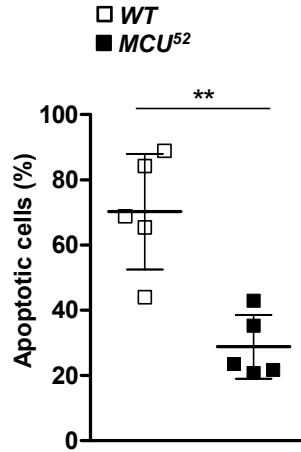


Figure S5

