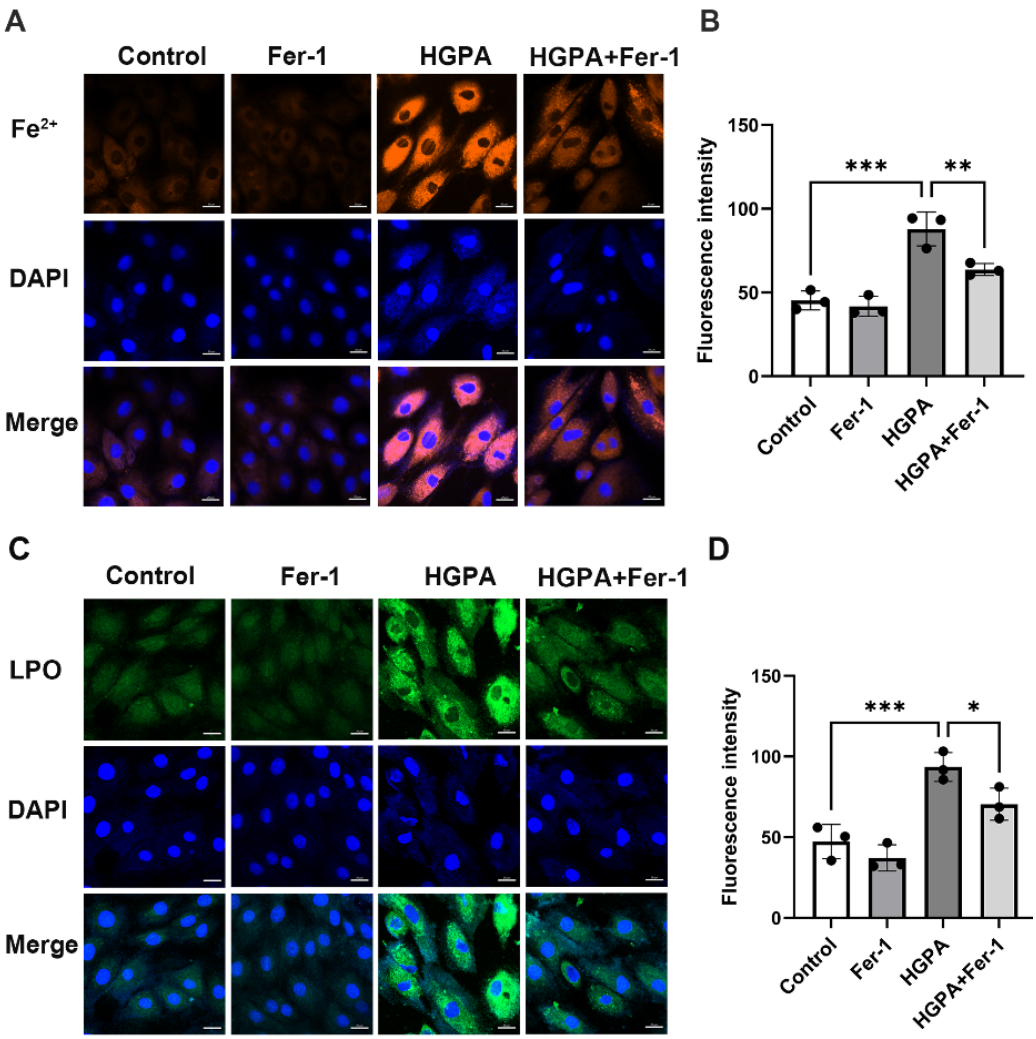
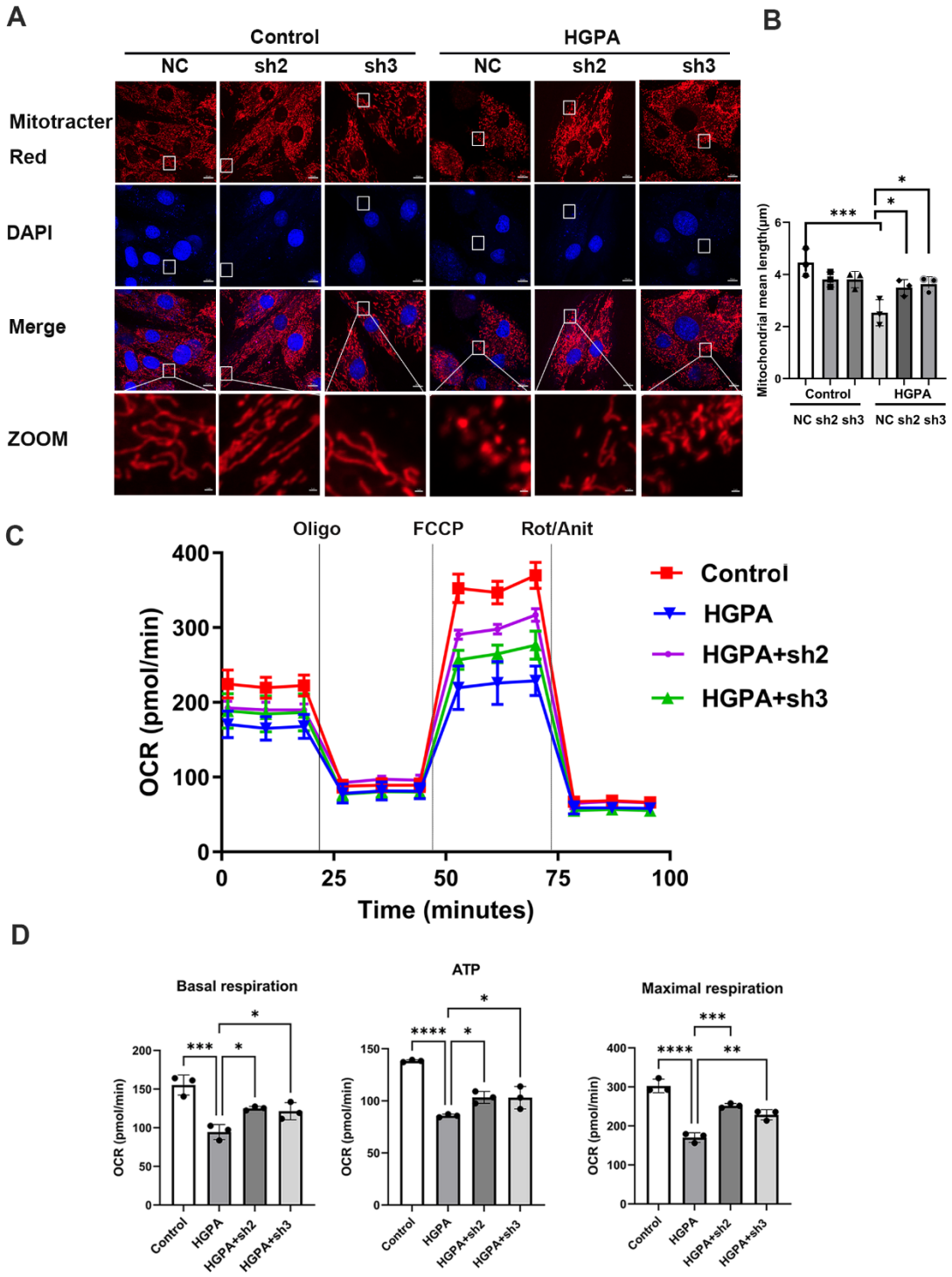


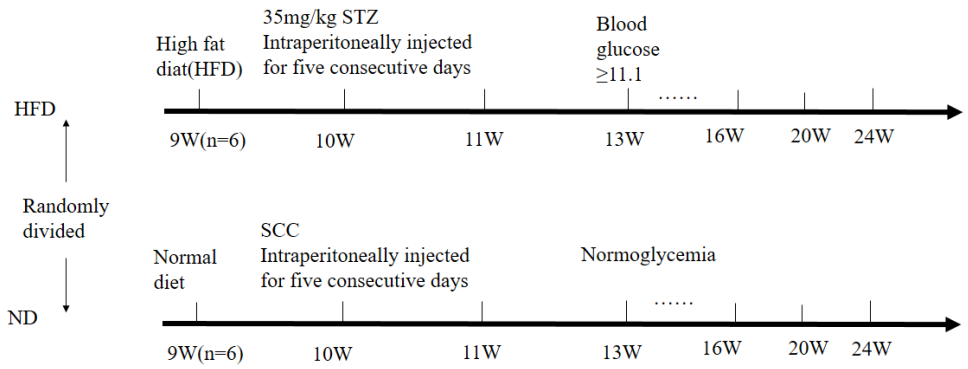
Supplementary Figure S1: Ferroptosis inhibitor reverses ferroptosis in cardiomyocytes.

(A) Effect of Fer-1 (10 μM added 2h before HGPA treatment) on iron ions in cardiomyocytes as detected by a fluorescent probe(n=3). Scale bar = 20 μm. (B) Effect of Fer-1 (10 μM added 2 hours before HGPA treatment) on lipid peroxidation in cardiomyocytes as detected by a fluorescent probe (n=3). Scale bar = 20 μm. Graphs show mean ± SD. statistical significance was analyzed using one-way ANOVA. *$P < 0.05$, **$P < 0.01$, ***$p < 0.001$.



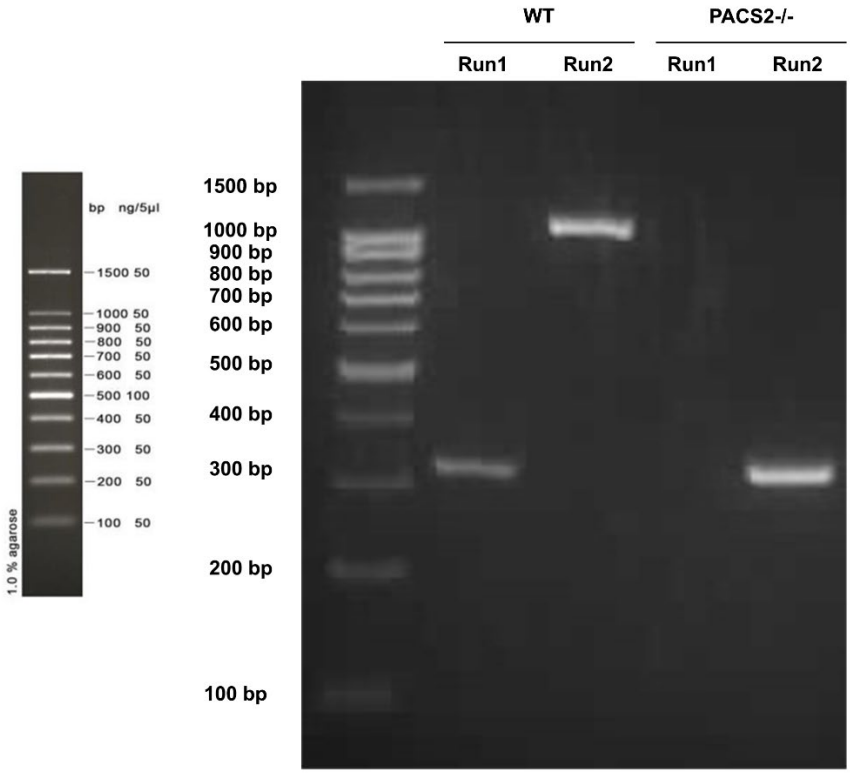
Supplementary Figure S2: Mitochondrial fragmentation and mitochondrial function.
(A,B) The length of each group of mitochondria was examined using confocal microscopy with a mitochondrial tracker(n=3). Scale bar = 63μm. (C,D)The mitochondrial stress test was used to measure the oxygen consumption rate (OCR) of the cells, and parameters reflecting mitochondrial function, basal respiration, ATP-coupled respiration, and maximal respiration were obtained(n=3). Graphs show mean ± SD. statistical significance was analyzed using one-way ANOVA. *$P < 0.05$, **$P < 0.01$, ***$p < 0.001$,****$p < 0.0001$.

A



Supplementary Figure S3: Mouse modeling of diabetes mellitus.

After 1 week of acclimatization, the mice were randomly divided into WT, PACS2$^{-/-}$, HFD, and HFD+PACS2$^{-/-}$ groups of 6 mice each, with the WT and PACS2$^{-/-}$ groups fed a standard diet and the HFD and HFD+PACS2$^{-/-}$ groups fed a HFD (D12109C, Research Diets, New Brunswick, NJ, USA); the latter two HFD groups were given 5 consecutive intraperitoneal injections of 35 mg/kg STZ (S0130, Sigma–Aldrich, San Louis, MO, USA) between 9 and 10 weeks. STZ was dissolved in sodium citrate-hydrochloric acid buffer (SSC) at pH 4.5. The WT and PACS2$^{-/-}$ groups were injected with the same volume of SSC. Blood glucose was measured at 14 weeks, and mice with blood glucose >11.1 mM were considered diabetic.



Supplementary Figure S4: Genotyping of PACS2$^{-/-}$ mice.

PACS2 genotypes were determined by PCR using the following primers:

P1- PACS2 (5'- TGCCCATAAGTAGGCAAAGCTTG-3')

P2- PACS2 (5'- TAAGGTAATAAACAGTTGCCCCCTT -3')

P3- PACS2 (5'- TGCCCATAAGTAGGCAAAGCTTG -3')

P4- PACS2 (5'- TCCCTAGGAGAGGCAAGGACA -3')

P1 and P2 produce a 323bp fragment of the wild-type (WT) locus.

P3 and P4 produced a 1099 bp fragment of the wild-type (WT) locus and a 328 bp fragment of the mutant locus.