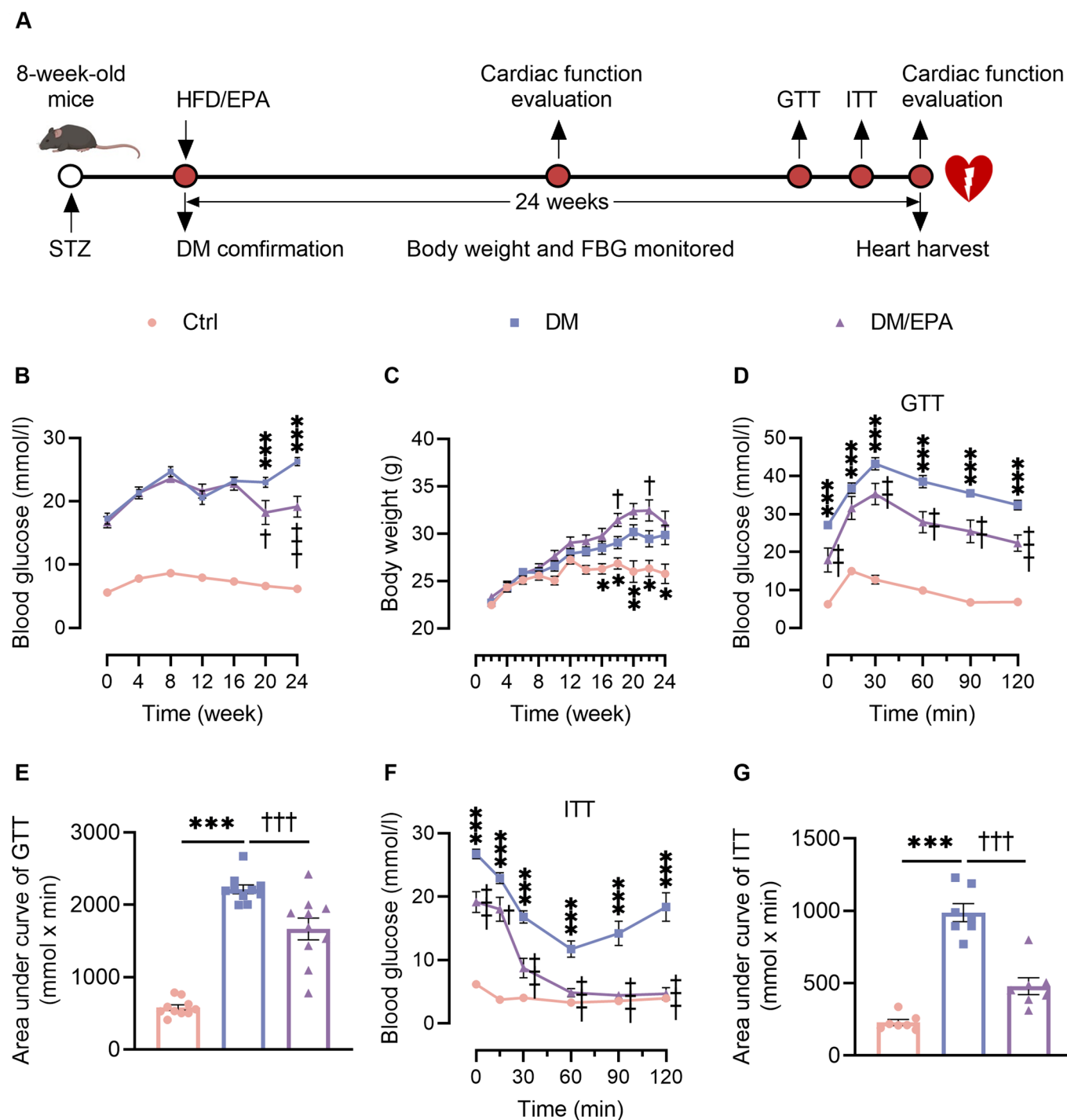
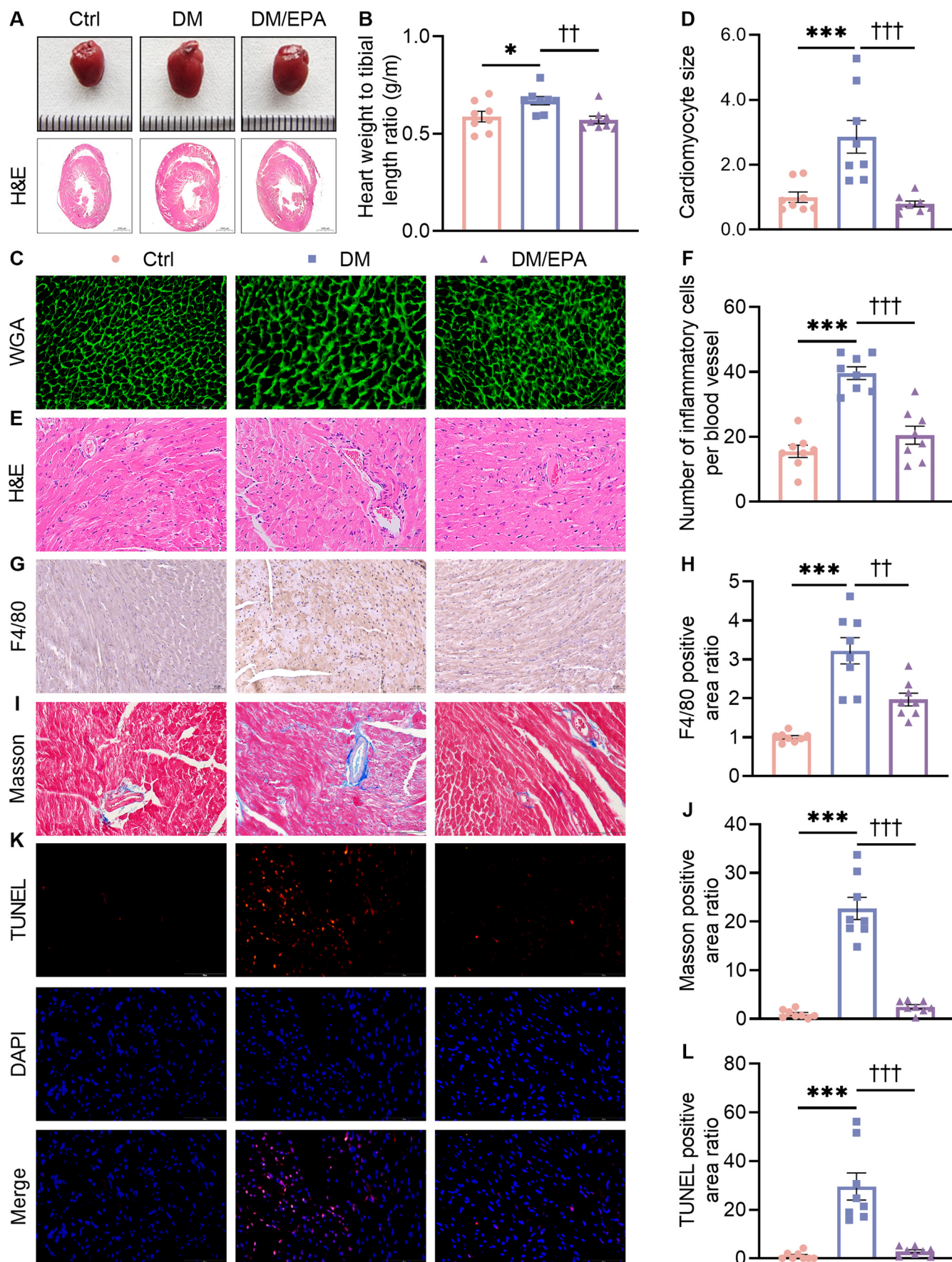


Expanded View Figures

Figure EV1. EPA lowered fasting blood glucose level, and improved glucose tolerance and insulin sensitivity in the diabetic mice.

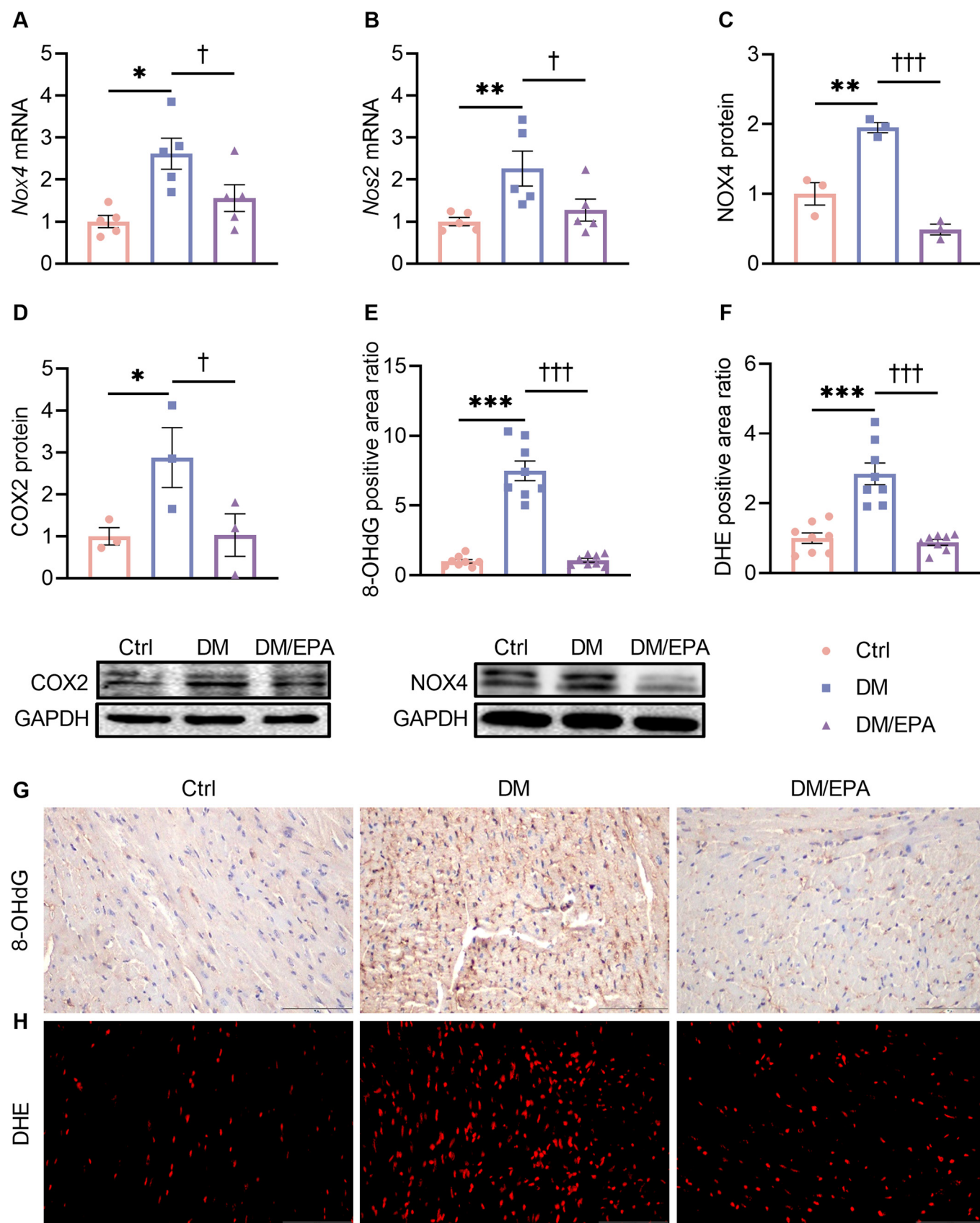
(A) Schematic diagram for the induction of DM and EPA treatment. (B) Fasting blood glucose levels. At the 20th week, *** $P < 0.001$, DM vs. Ctrl; [†] $P = 0.015$, DM/EPA vs. DM. At the 24th week, *** $P < 0.001$, DM vs. Ctrl; ^{†††} $P < 0.001$, DM/EPA vs. DM. (C) Body weight of the mice. Body weight was recorded every week, and presented every 2 weeks. At the 16th week, * $P = 0.044$, DM vs. Ctrl. At the 18th week, * $P = 0.027$, DM vs. Ctrl; [†] $P = 0.011$, DM/EPA vs. DM. At the 20th week, ** $P = 0.003$, DM vs. Ctrl. At the 22th week, * $P = 0.041$, DM vs. Ctrl; [†] $P = 0.032$, DM/EPA vs. DM. At the 24th week, * $P = 0.026$, DM vs. Ctrl. (D, E) Curve of blood glucose levels during GTT, and area under the curve. For (D), at the start, *** $P < 0.001$, DM vs. Ctrl; ^{††} $P = 0.002$, DM/EPA vs. DM. At the 15th minute, *** $P < 0.001$, DM vs. Ctrl. At the 30th minute, *** $P < 0.001$, DM vs. Ctrl; ^{††} $P = 0.009$, DM/EPA vs. DM. At the 60th minute, *** $P < 0.001$, DM vs. Ctrl; ^{††} $P = 0.001$, DM/EPA vs. DM. At the 90th minute, *** $P < 0.001$, DM vs. Ctrl; ^{††} $P = 0.001$, DM/EPA vs. DM. At the 120th minute, *** $P < 0.001$, DM vs. Ctrl; ^{†††} $P < 0.001$, DM/EPA vs. DM. For (E), *** $P < 0.001$, DM vs. Ctrl; ^{†††} $P < 0.001$, DM/EPA vs. DM. (F, G) Curve of blood glucose levels during ITT, and area under the curve. For (F), at the start, *** $P < 0.001$, DM vs. Ctrl; ^{†††} $P < 0.001$, DM/EPA vs. DM. At the 15th minute, *** $P < 0.001$, DM vs. Ctrl. [†] $P = 0.01$, DM/EPA vs. DM. At the 30th, 60th, 90th, and 120th minute, *** $P < 0.001$, DM vs. Ctrl; ^{†††} $P < 0.001$, DM/EPA vs. DM. For (G), *** $P < 0.001$, DM vs. Ctrl; ^{†††} $P < 0.001$, DM/EPA vs. DM. Data information: Data are represented as individual data points of $n = 10, 14, 14$ (B), $n = 10, 14, 14$ (C), $n = 10, 10, 10$ (D, E), $n = 7, 7, 7$ (F, G) biological replicates and means $\pm$ SEM. Analysis by one-way ANOVA. AUC area under curve, EPA eicosapentaenoic acid, FBG fasting blood glucose, GTT glucose tolerance test, HFD high-fat diet, ITT insulin tolerance test, STZ streptozotocin. Groups: Ctrl control, DM diabetes mellitus, DM/EPA diabetic mice supplemented with EPA.





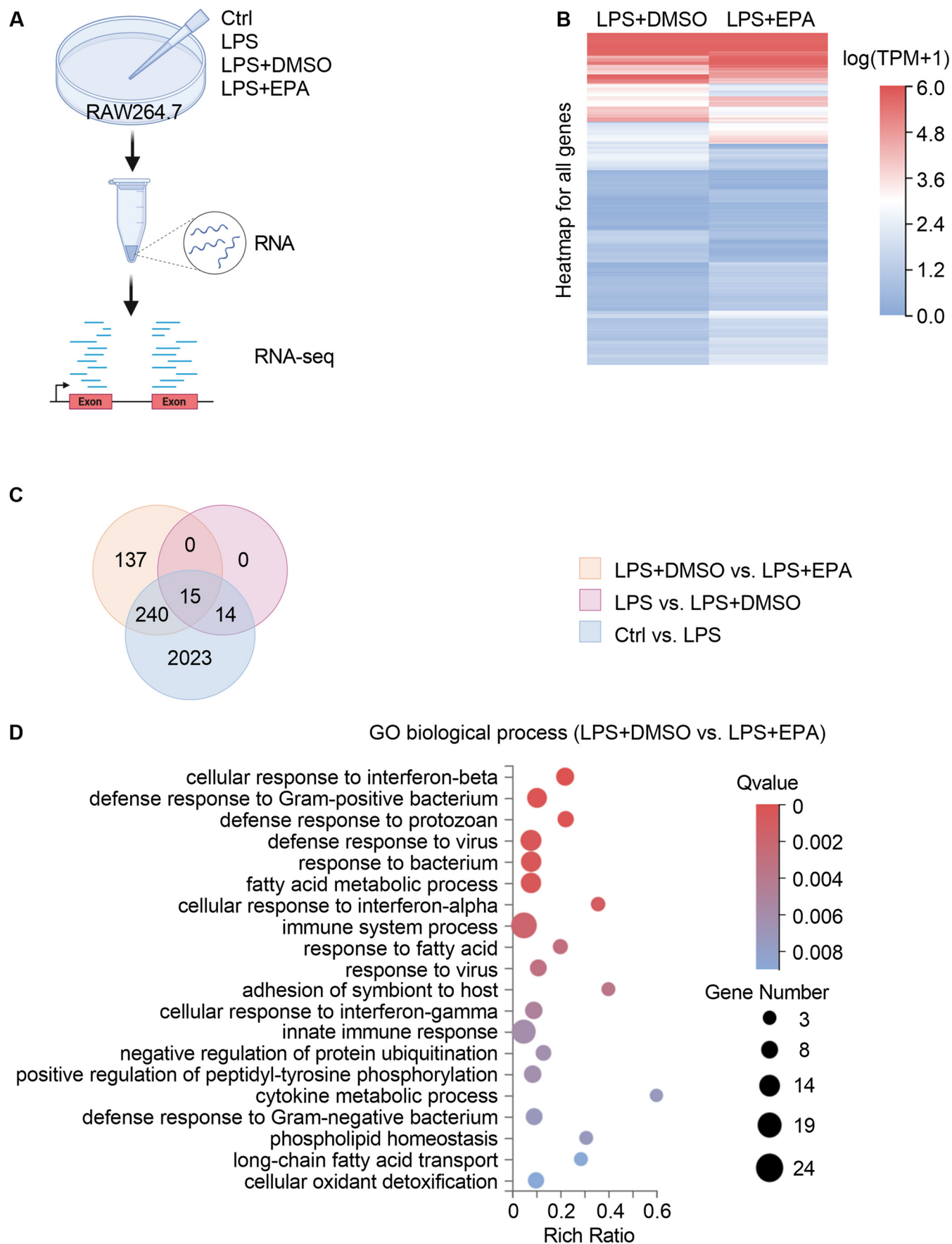
◀ **Figure EV2. EPA prevented DM-induced cardiac pathological injuries.**

(A) Representative gross images of the entire hearts (upper panel, bar = 1 mm), and cross-section images of H&E staining of the hearts (lower panel, bar = 1000 μ m). (B) The ratio of heart-weight-to-tibia length. * $P = 0.021$, DM vs. Ctrl; $^{††}P = 0.006$, DM/EPA vs. DM. (C, D) WGA staining (bar = 100 μ m) and quantification of cardiac hypertrophy. For (D), *** $P < 0.001$, DM vs. Ctrl; $^{†††}P < 0.001$, DM/EPA vs. DM. (E, F) H&E staining (bar = 100 μ m) and the number of inflammatory cells infiltrated per vessel. For (F), *** $P < 0.001$, DM vs. Ctrl; $^{†††}P < 0.001$, DM/EPA vs. DM. (G, H) Immunohistochemical staining of F4/80 and quantification of positive area (bar = 50 μ m). For (H), *** $P < 0.001$, DM vs. Ctrl; $^{††}P = 0.001$, DM/EPA vs. DM. (I, J) Masson's trichrome staining (bar = 100 μ m) with the positive area quantified. For (J), *** $P < 0.001$, DM vs. Ctrl; $^{†††}P < 0.001$, DM/EPA vs. DM. (K, L) TUNEL assay (bar = 100 μ m) by double staining with DAPI (blue) and TUNEL (red), and ratio of the TUNEL positive nuclei. For (L), *** $P < 0.001$, DM vs. Ctrl; $^{†††}P < 0.001$, DM/EPA vs. DM. Data information: For (D, H, J, L), the data are normalized to Ctrl. Data are represented as individual data points of $n = 8$ (B, D, F, H, J, L) biological replicates and means $\pm$ SEM. Analysis by one-way ANOVA. DAPI 4',6-diamidino-2-phenylindole, TUNEL terminal deoxynucleotidyl transferase (TdT)-mediated dUTP nick-end labeling, WGA wheat germ agglutinin. Other abbreviations are the same as in Fig. EV1. Groups: Ctrl control, DM diabetes mellitus, DM/EPA diabetic mice supplemented with EPA.



◀ **Figure EV3. EPA attenuated DM-induced cardiac oxidative stress.**

(A, B) mRNA expression of *Nox4* and *Nos2* were determined by qRT-PCR. For (A), $^*P = 0.02$, DM vs. Ctrl; $^{\dagger}P = 0.025$, DM/EPA vs. DM. For (B), $^{**}P = 0.009$, DM vs. Ctrl; $^{\dagger}P = 0.032$, DM/EPA vs. DM. (C, D) Protein levels of NOX4 and COX2 were determined by Western blot. For (C), $^{**}P = 0.001$, DM vs. Ctrl; $^{+++}P < 0.001$, DM/EPA vs. DM. For (D), $^*P = 0.043$, DM vs. Ctrl; $^{\dagger}P = 0.045$, DM/EPA vs. DM. (E) Quantification of 8-OHdG positive area of the hearts. $^{***}P < 0.001$, DM vs. Ctrl; $^{+++}P < 0.001$, DM/EPA vs. DM. (F) Quantification of DHE positive area of the hearts. $^{***}P < 0.001$, DM vs. Ctrl; $^{+++}P < 0.001$, DM/EPA vs. DM. (G) Immunohistochemical staining of 8-OHdG (bar = 100 μm). (H) DHE staining of heart sections (bar = 100 μm). Data information: For (A–F), the data are normalized to Ctrl. Data are represented as individual data points of $n = 5, 5, 5$ (A, B), $n = 3, 3, 3$ (C, D), $n = 8, 8, 8$ (E), $n = 8, 8, 8$ (F) biological replicates and means $\pm$ SEM. Analysis by one-way ANOVA. 8-OHdG 8-hydroxy-2'-deoxyguanosine, COX2 cyclooxygenase-2, DHE dihydroethidium, *Nos2* nitric oxide synthase 2, *Nox4*/NOX4 NADPH oxidase 4. Other abbreviations are the same as in Fig. EV1. Groups: Ctrl control, DM diabetes mellitus, DM/EPA diabetic mice supplemented with EPA.



**Figure EV4. EPA regulated immune-associated signal pathways in M1-polarized macrophages.**

(A) Schematic representation of the RNA-seq protocol. (B) Heat map comparison of the global mRNA expression profiles in LPS + DMSO and LPS + EPA groups. (C) Venn diagram showing comparisons of altered gene numbers among the groups. (D) GO analysis for top biological processes altered by EPA compared with DMSO in LPS-challenged RAW264.7 cells. GO Gene Ontology, LPS lipopolysaccharides, RNA-seq RNA-sequencing. Other abbreviations are the same as in Fig. EV1. Groups: Ctrl control, LPS LPS-stimulated RAW264.7 cells, LPS + DMSO LPS-stimulated RAW264.7 cells treated with DMSO, LPS + EPA LPS-stimulated RAW264.7 cells treated with EPA.

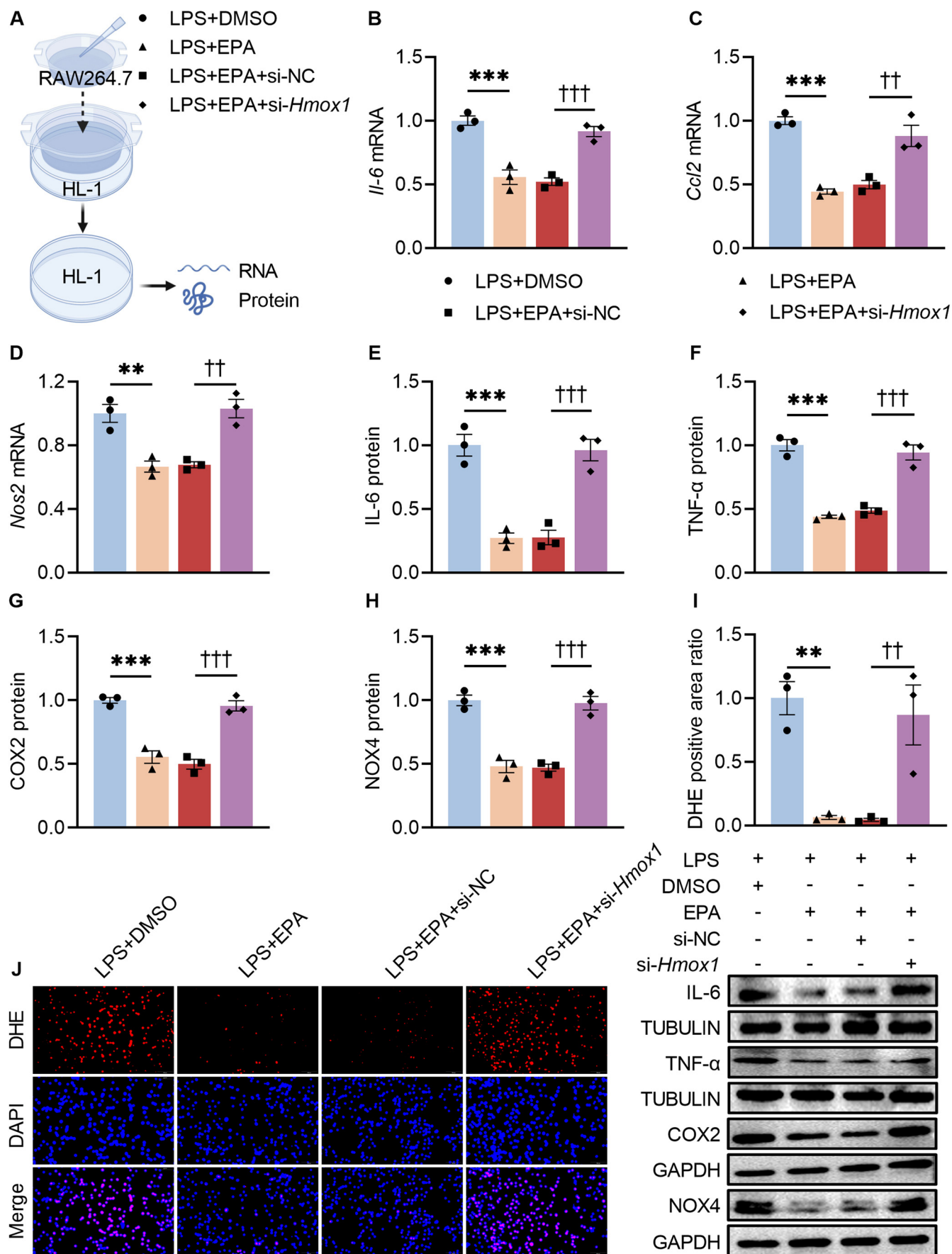


Figure EV5. HO-1 was required for EPA repression of cardiomyocyte injury induced by M1 polarized macrophages.

(A) Schematic representation of the experimental protocol. (B–D) mRNA levels of *Il-6*, *Ccl2*, and *Nos2* in HL-1 cells. For (B), *** $P < 0.001$, LPS + EPA vs. LPS + DMSO; ^{†††} $P < 0.001$, LPS + EPA+si-*Hmox1* vs. LPS + EPA+si-NC. For (C), *** $P < 0.001$, LPS + EPA vs. LPS + DMSO; ^{††} $P = 0.001$, LPS + EPA+si-*Hmox1* vs. LPS + EPA+si-NC. For (D), ** $P = 0.001$, LPS + EPA vs. LPS + DMSO; ^{††} $P = 0.001$, LPS + EPA+si-*Hmox1* vs. LPS + EPA+si-NC. (E–H) Protein levels of IL-6, TNF- α , COX2, and NOX4. *** $P < 0.001$, LPS + EPA vs. LPS + DMSO; ^{†††} $P < 0.001$, LPS + EPA+si-*Hmox1* vs. LPS + EPA+si-NC. (I, J) DHE assay (bar = 100 μ m) by double staining with DAPI (blue) and DHE (red), and ratio of DHE positive cells. For (I), ** $P = 0.001$, LPS + EPA vs. LPS + DMSO; ^{††} $P = 0.003$, LPS + EPA+si-*Hmox1* vs. LPS + EPA+si-NC. Data information: For (B–I), the data are normalized to LPS + DMSO. Data are represented as individual data points of $n = 3$ (B–I) biological replicates and means $\pm$ SEM. Analysis by one-way ANOVA. *Ccl2* C-C motif chemokine ligand 2, *Il-6*/IL-6 interleukin-6, si-*Hmox1* *Hmox1* siRNA, si-NC negative control siRNA, TNF- α tumor necrosis factor- α . Other abbreviations are the same as in Figs. EV1–EV4. Groups: LPS + DMSO HL-1 cells co-cultured with LPS-stimulated RAW264.7 cells and treated with DMSO, LPS + EPA HL-1 cells co-cultured with LPS-stimulated RAW264.7 cells and treated with EPA, LPS + EPA+si-NC LPS + EPA treated with si-NC, LPS + EPA+si-*Hmox1* LPS + EPA treated with si-*Hmox1*.