Supplemental data

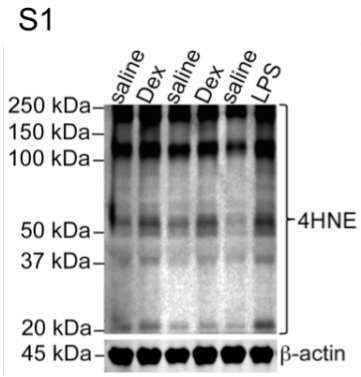

Fig. S1. Dexamethasone injection induces oxidative stress in white vastus muscles in mice. Western blot was performed for 4-HNE in muscle homogenates in mice i.p. injection of Dex (25 mg/kg) for 7 days. A sample from a mouse with lipopolysaccharide (LPS: 1 mg/kg, 12 hrs) injection were used as positive control.

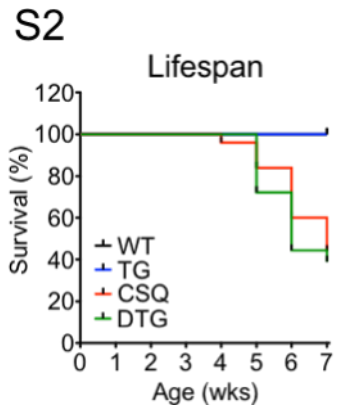

Fig. S2. Muscle-specific overexpression of EcSOD does not improve survival in mice with CHF. Kaplan–Meier survival curves for MCK-EcSOD (TG), α-MHC-CSQ (CSQ) and MCK-EcSOD:α-MHC-CSQ double transgenic mice (DTG) and their wild-type littermates (WT). Death occurred between 5-7 wks of age and showed no significant differences between CSQ and DTG mice (n=12-25/group).

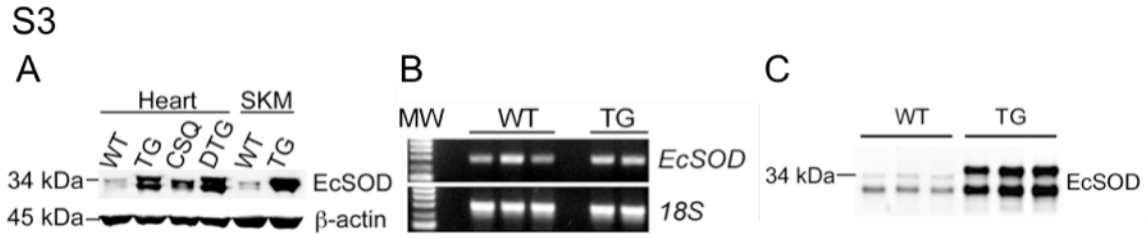

Fig. S3. Elevated EcSOD levels in the heart of muscle-specific EcSOD transgenic mice due to redistribution through circulation. A) Western blot image showing significantly increased EcSOD protein expression in the hearts of MCK-EcSOD mice (TG) compared with wild-type littermates (WT). WT and TG plantaris skeletal muscle (SKM) were used as controls; B) Semi-quantitative RT-PCR showing similar EcSOD mRNA expression in both TG and WT mice; and C) Western blot image showing significantly elevated EcSOD levels in the serum of TG mice compared with WT mice.

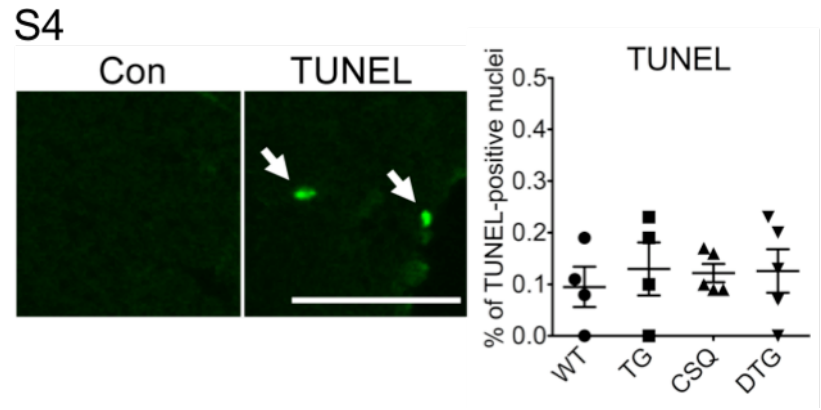

Fig. S4. Measurement of apoptosis in skeletal muscle. Fresh frozen plantaris sections were used to detect apoptosis by TUNEL staining with DAPI staining for nuclei staining. The number of TUNEL-positive nuclei was counted from at least 1000 nuclei. Representative TUNEL staining (left, scale bar = 50 μm) and quantification of the apoptosis (right) at 7 wks of age (n=4-6/group).

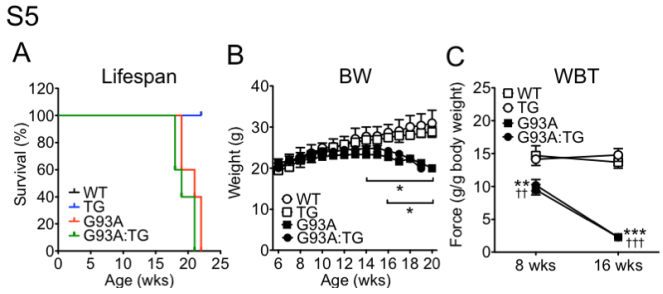

Fig. S5. Muscle-specific overexpression of EcSOD does not reduce mortality, loss of body weight and muscle function in ALS mice. MCK-EcSOD mice (TG) were crossbred with SOD1-G93A mice

(G93A) to generate G93A:TG double transgenic mice and measured for life span, body weight (BW) and whole body tension test (WBT). A) Kaplan–Meier survival curves for TG, G93A, G93A:TG and WT mice. Death occurred between 20 wks of age and showed no significant differences between G93A and G93A:TG mice (n=4-5/group); B) Body weight in G93A and G93A:TG mice were significantly decreased starting at 14 wks of age. * denotes $p<0.05$; C) Muscle force production measured by WBT (n=4-5/group). ** and *** denote $p<0.01$ and $p<0.001$, respectively, between G93A and WT at either 8 or 16 wks. †† and ††† denote $p<0.01$ and $p<0.001$, respectively, between G93A:TG and TG at either 8 or 16 wks.

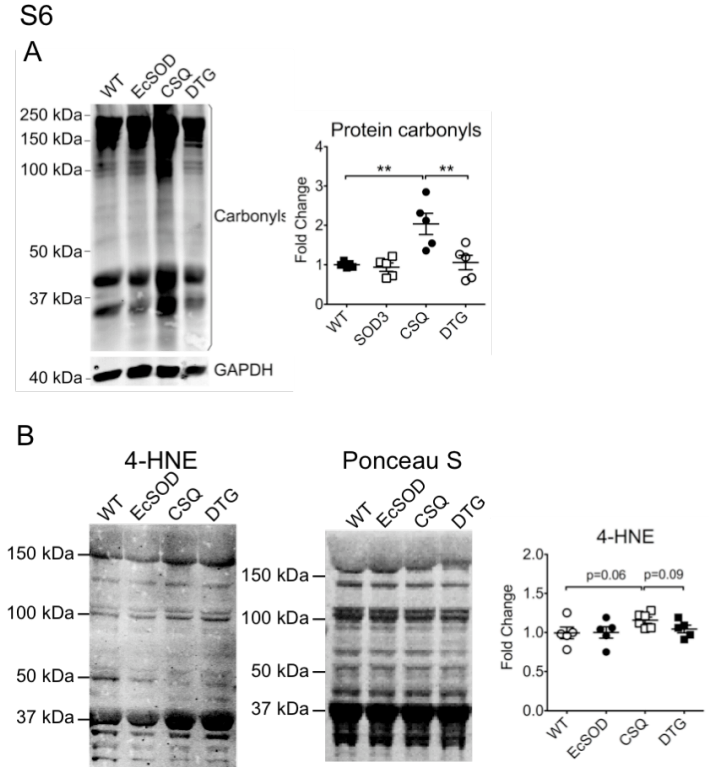

Fig. S6. Muscle-specific overexpression of EcSOD prevents CHF-induced oxidative stress in skeletal muscle. Glycolytic white vastus lateralis muscles (WV) were harvested from MCK-EcSOD (TG), α-MHC-CSQ (CSQ) and MCK-EcSOD:α-MHC-CSQ double transgenic mice (DTG) and their wild-type littermates (WT) at 7 wks of age and processed for measurements of oxidative stress. A) Representative Western blot images of protein carbonylation in WV muscle with GAPDH as loading control. Quantification with statistical analysis were shown on the right; and B) Representative Western blot images of 4-HNE in WV muscle with Ponceau S staining as loading control. Quantification with statistical analysis were shown on the right. ** denotes $p<0.01$.