

SUPPLEMENTARY MATERIALS

Fig. S1. Expression of nNOS in nNOSOE myocytes.

Fig. S2. ROS levels in Sed/Ex WT and nNOSKO myocytes.

Fig. S3. Ex-nNOSKO myocytes have oxidative-stress induced contractile dysfunction.

Fig. S4. Effects of exogenous NO on contraction.

Fig. S5. Effects of EMEPO on contraction.

Fig. S6. Representative blots of PLB Serine16 phosphorylation.

Fig. S7. Effects of myocyte-specific nNOS overexpression on contraction.

Fig. S8. Exercise and overexpression of nNOS does not alter PLB and SERCA expression.

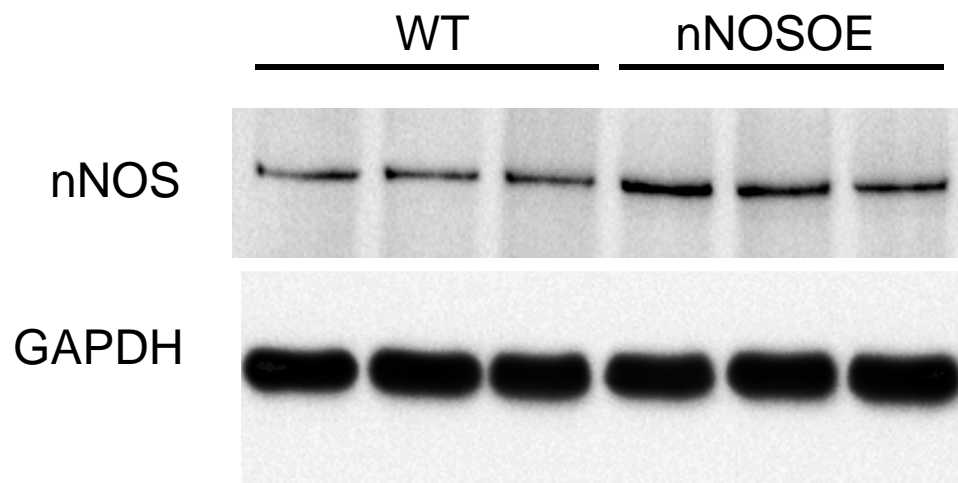


Figure S1. Increased expression of nNOS in nNOSOE myocytes.

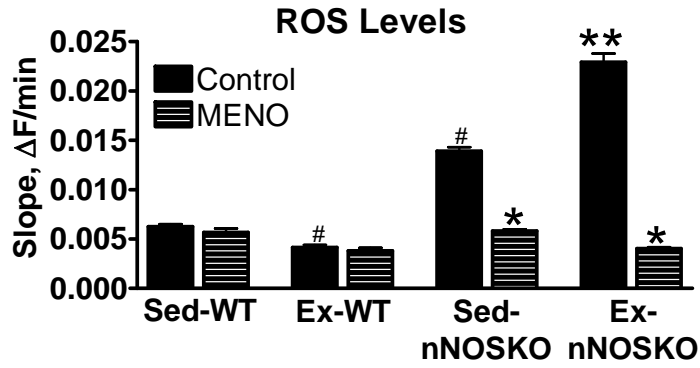


Figure S2. ROS levels (\pm $O_2^{\cdot-}$ scavenger MENO) in Sed/Ex WT and nNOSKO myocytes. Summary data for ROS levels (measured as slope) (\pm MENO), n=19-42 cells/3-4 hearts. # P<0.05 vs Sed-WT, * P<0.05 vs corresponding control, ** P<0.05 vs all other groups.

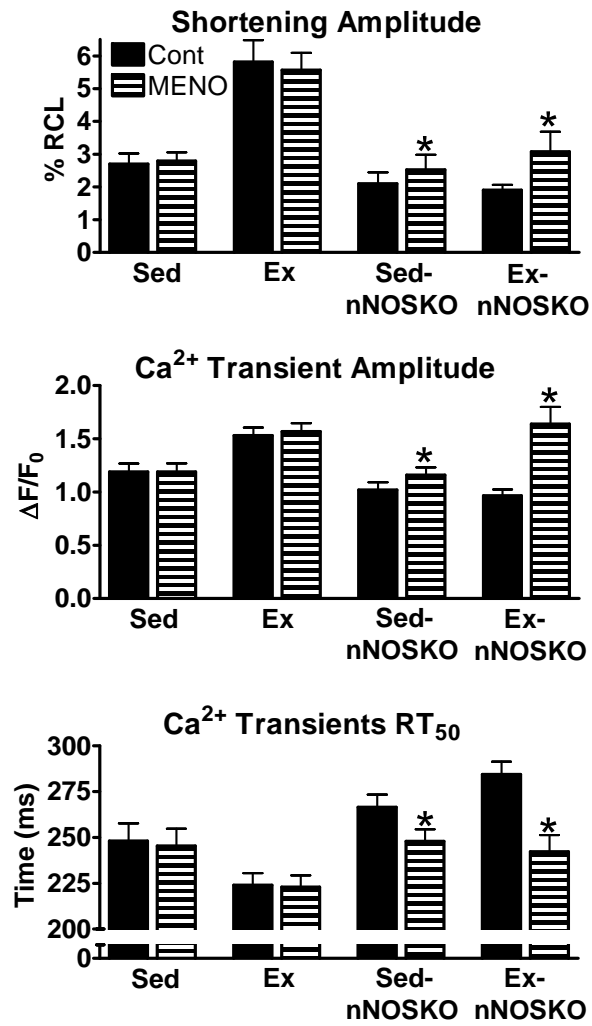


Figure S3. Exercise-trained nNOSKO myocytes have oxidative-stress induced contractile dysfunction. Summary data of shortening (top), Ca²⁺ transient amplitude (middle) and Ca²⁺ transient decline (measured as time to 50% relaxation-RT₅₀) (bottom), n=15-23 cells/4 hearts. * P<0.05 vs corresponding control.

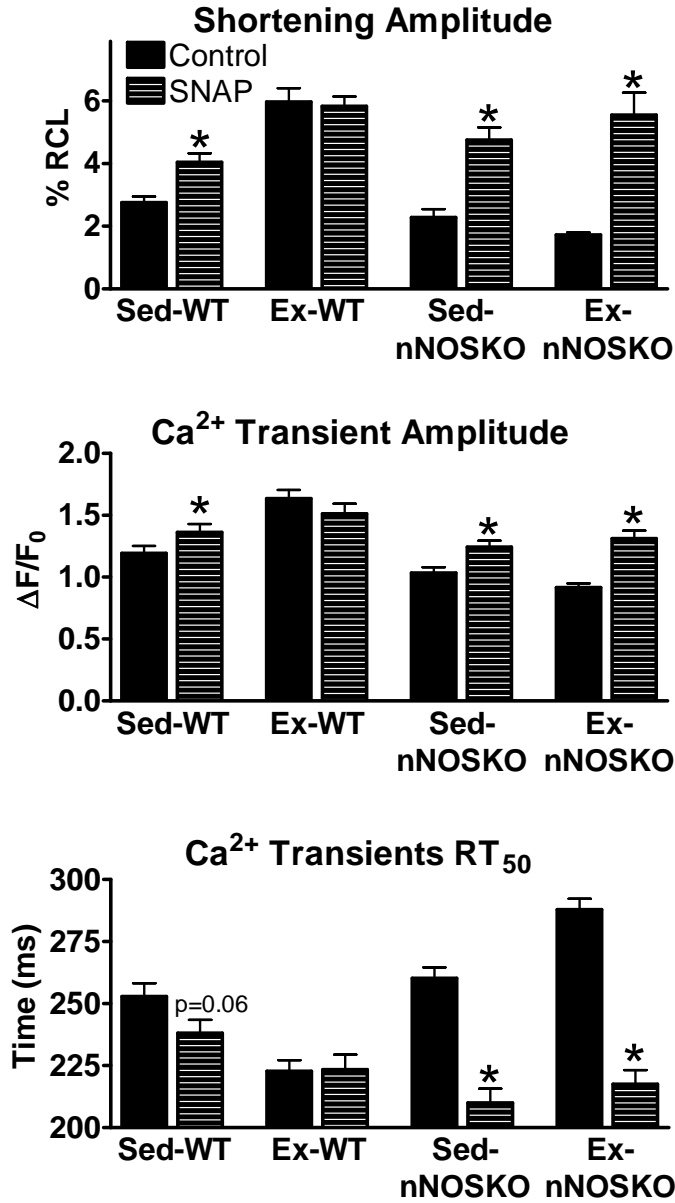


Figure S4. Effects of exogenous NO (SNAP) on contraction from WT and nNOSKO sedentary (Sed) and exercise-trained (Ex) myocytes. Summary data of shortening (top), Ca²⁺ transient amplitude (middle) and Ca²⁺ transient decline (measured as time to 50% relaxation-RT₅₀) (bottom), n=23-28 cells/4-5 hearts. * P<0.05 vs corresponding control.

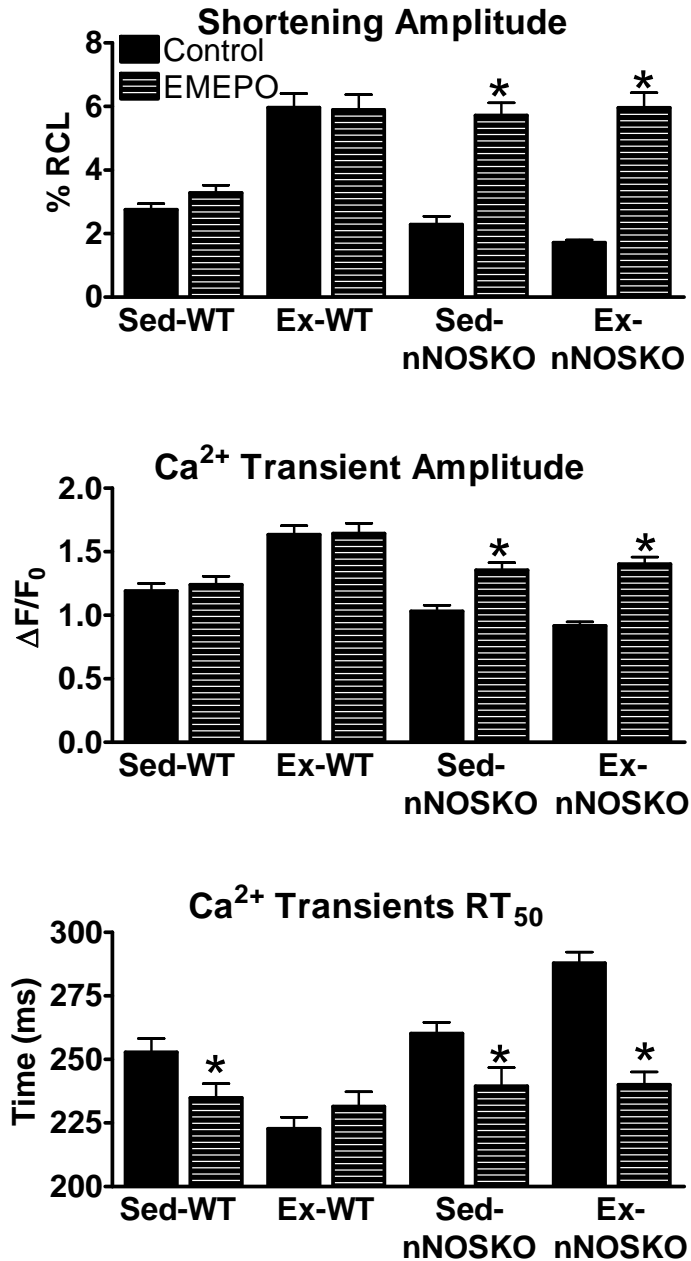


Figure S5. Effects of EMEPO on contraction from WT and nNOSKO sedentary (Sed) and exercise-trained (Ex) myocytes. Summary data of shortening (top), Ca²⁺ transient amplitudes (middle) and Ca²⁺ transient decline (measured as time to 50% relaxation-RT₅₀) (bottom), n=19-34 cells/4-5 hearts. * P<0.05 vs corresponding control.

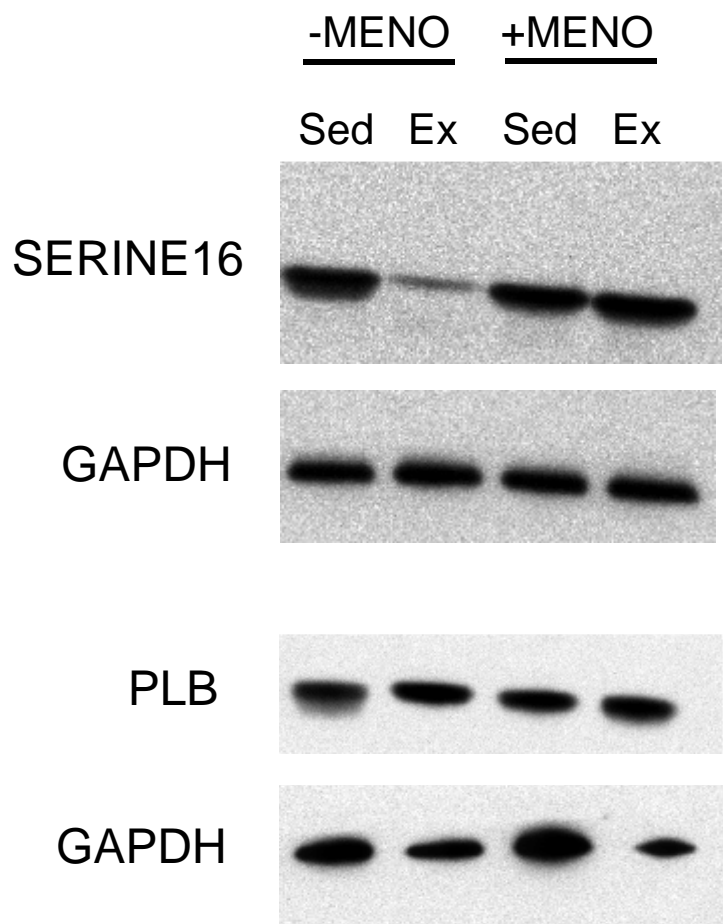


FIGURE S6. Representative blots of PLB total and Serine16 phosphorylation (and GAPDH) in Sed and Ex trained nNOSKO myocytes.

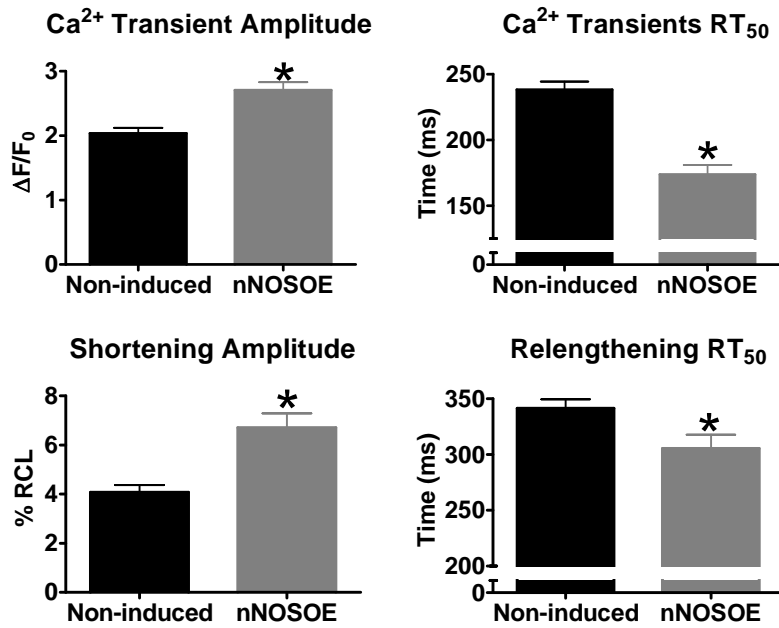


Figure S7. Effects of myocyte-specific nNOS overexpression (nNOSOE) on contraction. Summary data of Ca²⁺ transient (top) and shortening (bottom) and relaxation (right) (measured as time to 50% relaxation-RT₅₀), n=24-34 cells/6 hearts. * P<0.05 vs control.

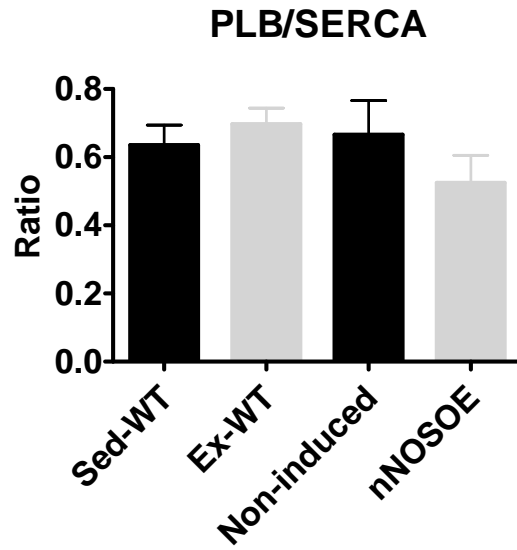


Figure S8. Exercise and overexpression of nNOS does not alter PLB and SERCA expression. Summary data of the PLB/SERCA ratio (PLB/SERCA expression normalized to GAPDH). n=6 hearts/group. P=NS.