

Supplementary data to the article:

S-nitrosoglutathione preserves vasodilation and attenuates myocardial ischemia-reperfusion injury

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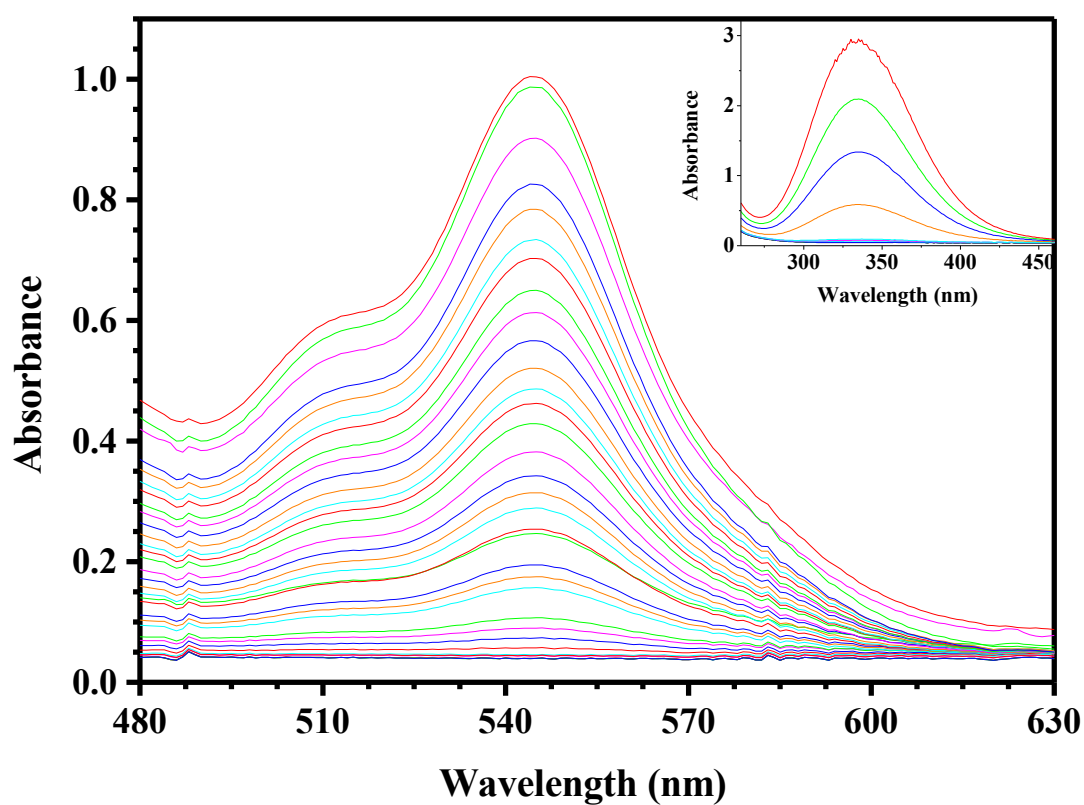


Figure S1. Spectral change during the thermal decomposition reaction of GSNO with NO release, showing the decay of characteristic absorption bands with maxima at 545 nm ($n \rightarrow \pi^*$ transition, $\epsilon = 15.9 \text{ M}^{-1} \text{ cm}^{-1}$) and 336 nm ($\pi \rightarrow \pi^*$ transition, $\epsilon = 922 \text{ M}^{-1} \text{ cm}^{-1}$). The shoulder with a maximum at ca. 515 nm is attributed to the absorption band of the syn conformer, which is superimposed on the more intense band of the anti conformer.

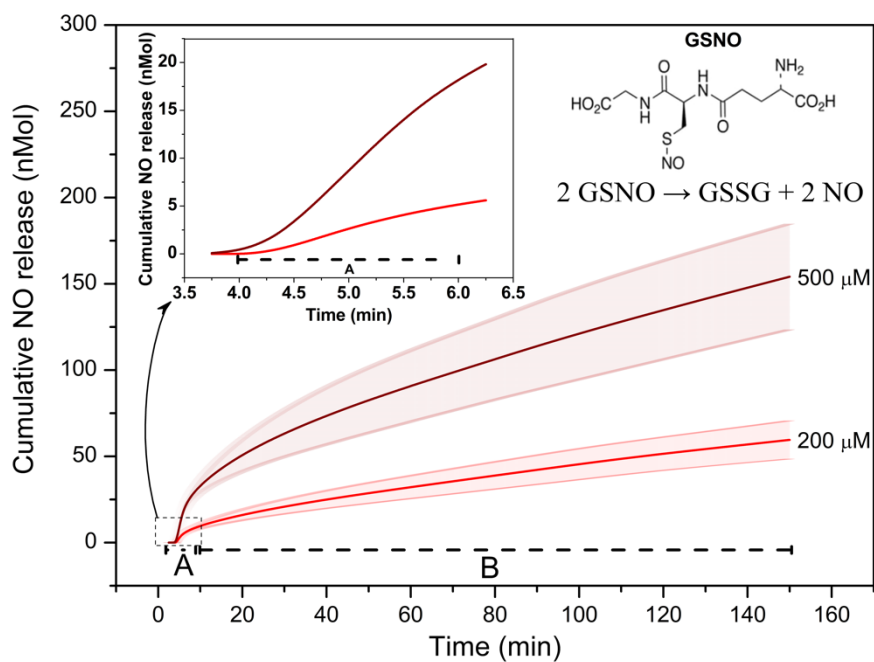


Figure S2. Cumulative nitric oxide (NO) release from KH-GSNO solutions at 200 μM and 500 μM measured by ozone-based chemiluminescence. Cumulative NO release was monitored over 150 min at 37 °C. The inset shows the initial release behavior immediately after GSNO injection. Two kinetic regimes were quantified: Region A, corresponding to the rapid initial burst, and Region B, representing the subsequent slow-decay phase. Release rates for 200 μM GSNO were 28.80 and 3.45 $\text{nMol}\cdot\text{mL}^{-1}\cdot\text{min}^{-1}$ in Regions A and B, respectively, whereas the 500 μM solution exhibited rates of 97.78 and 8.20 $\text{nMol}\cdot\text{mL}^{-1}\cdot\text{min}^{-1}$.

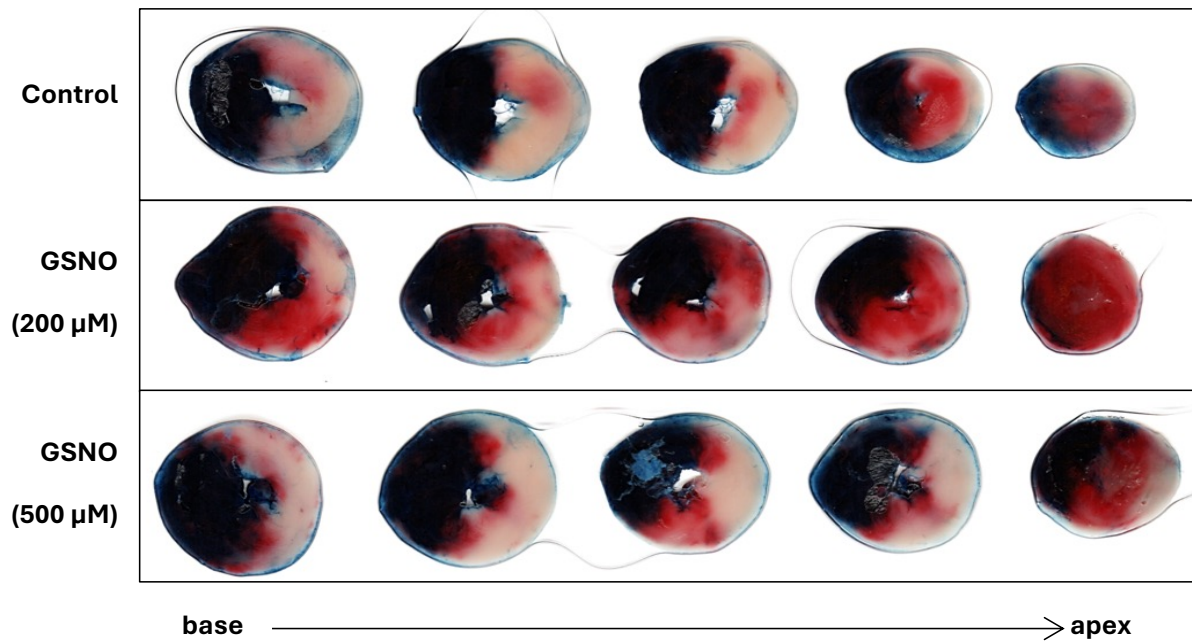
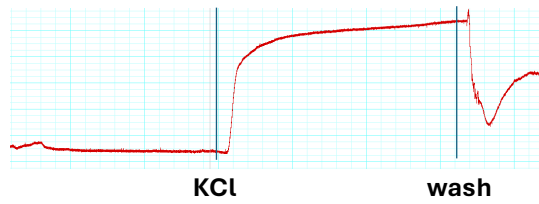


Figure S3. Representative TTC-stained myocardial slices from each experimental group. Following restoration of the ischemic region and Evans blue injection, hearts were sectioned into five transverse slices (2–3 mm). Evans blue delineates the non-ischemic myocardium (blue), TTC identifies viable myocardium within the area at risk (red), and infarcted myocardium is identified by the absence of staining (white).

a)

Smooth muscle viability assay
(KCl 60 mM, 15 min)



b)

Endothelial viability test
ACh-induced relaxation

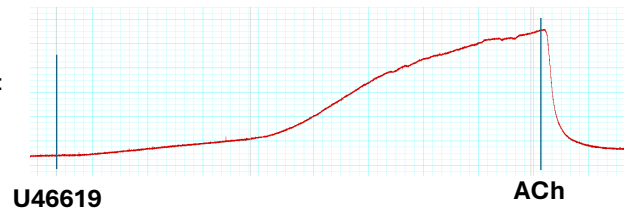


Figure S4. Vascular reactivity traces illustrating a) smooth muscle viability (KCl-induced contraction) and b) endothelial function (acetylcholine-induced relaxation) in coronary artery rings.

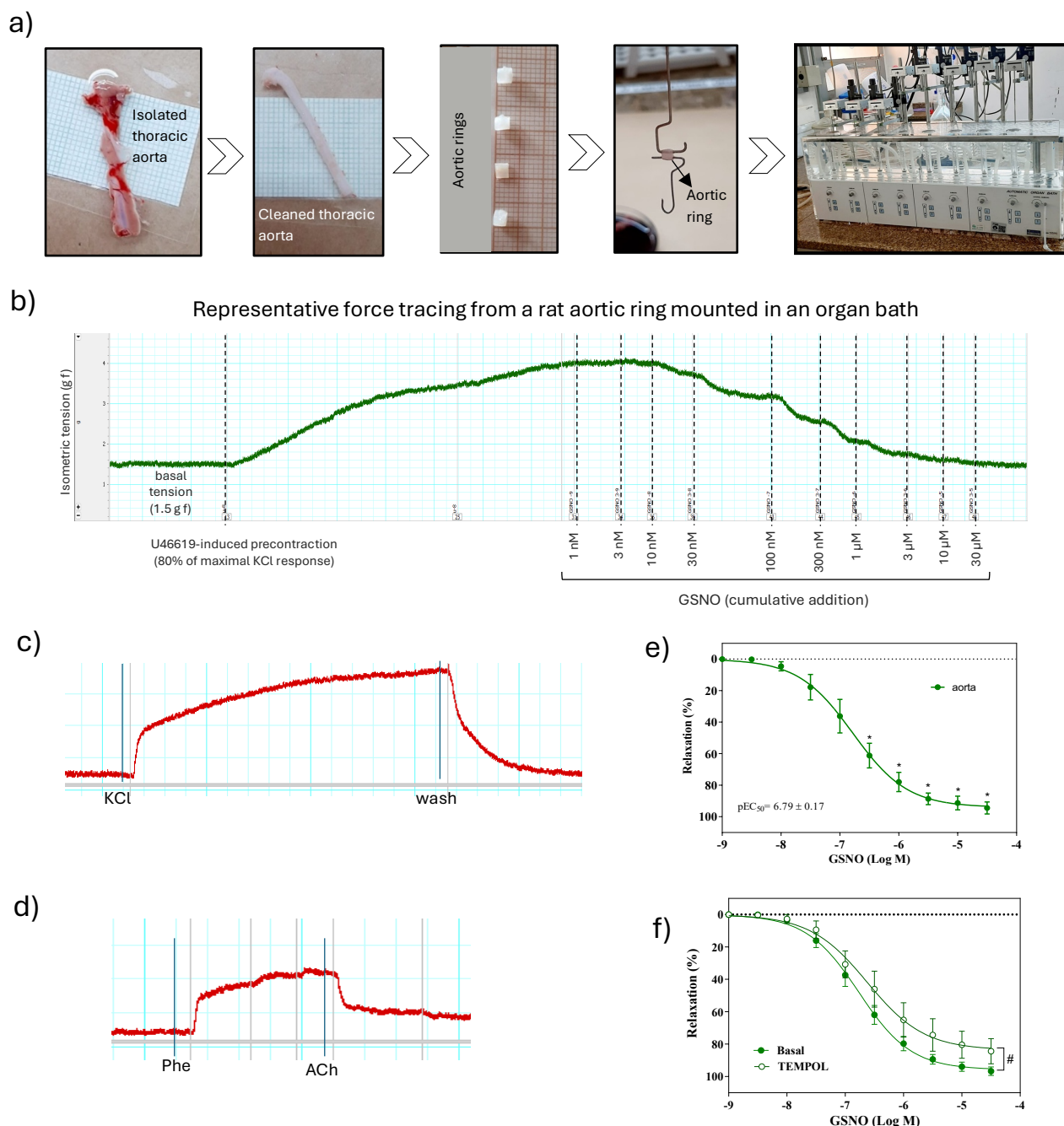


Figure S5. a) Representative images showing the isolation of the aorta for mounting the rings in an organ bath. b) Representative traces showing the responses of aorta rings to cumulative concentrations of GSNO (1 nM–30 μ M). Vascular reactivity traces illustrating c) smooth muscle viability (KCl-induced contraction) and d) endothelial function (acetylcholine-induced relaxation) in aorta rings. e) Concentration–response curve to GSNO in aorta rings. The pEC_{50} value (negative logarithm of the agonist concentration producing half-maximal response) is indicated within the graph. Data are presented as mean $\pm$ SEM ($n = 3$ /group). One-way ANOVA followed by Dunnett's multiple comparisons test: * $p < 0.05$ vs. -9 Log M . f) Concentration-response curves to GSNO in aorta rings in the absence (Basal) and presence of the superoxide scavenger TEMPOL (100 μ M) after a 30-min incubation. Data are presented as mean $\pm$ SEM ($n = 3$ /group). Two-way repeated measures ANOVA followed by Bonferroni's post hoc test: # $p < 0.05$ vs. Basal.

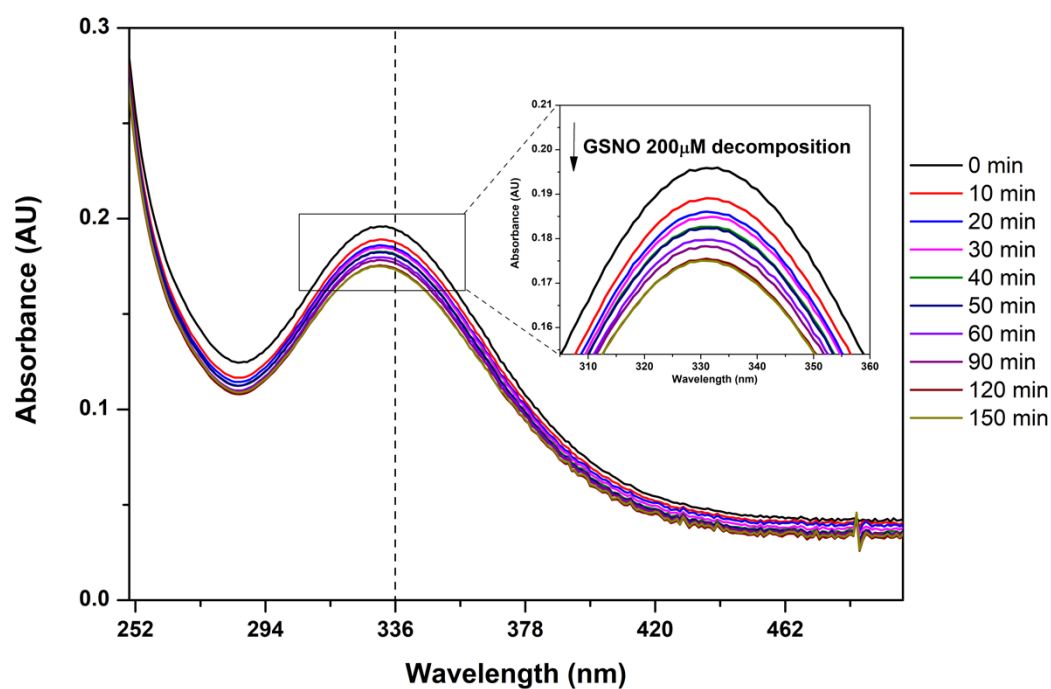


Figure S6. Time-dependent UV-Vis spectra of GSNO (200 μM) monitored over 150 min under physiological conditions. The decrease in absorbance at the characteristic S-NO $\pi \rightarrow \pi^*$ transition band (~ 336 nm) reflects the progressive photolytic and thermal decomposition of GSNO. Spectra were recorded at 0, 10, 20, 30, 40, 50, 60, 90, 120, and 150 min. The inset provides a magnified view of the 310–360 nm region, highlighting the gradual loss of absorbance associated with GSNO degradation. Together, these data illustrate the kinetic profile of GSNO decay within the studied time frame.

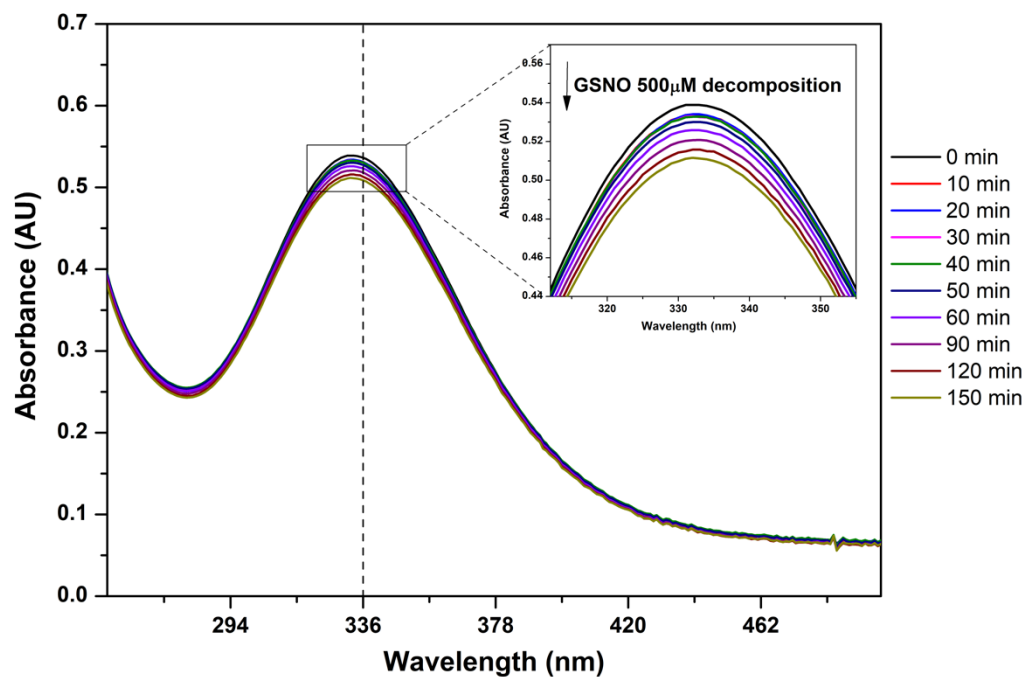


Figure S7. Time-dependent UV-Vis spectra of GSNO (500 μM) monitored over 150 min under physiological conditions. The decrease in absorbance at the characteristic S–NO $\pi\rightarrow\pi^*$ transition band (~ 336 nm) reflects the progressive photolytic and thermal decomposition of GSNO. Spectra were recorded at 0, 10, 20, 30, 40, 50, 60, 90, 120, and 150 min. The inset provides a magnified view of the 310–360 nm region, highlighting the gradual loss of absorbance associated with GSNO degradation. Together, these data illustrate the kinetic profile of GSNO decay within the studied time frame.