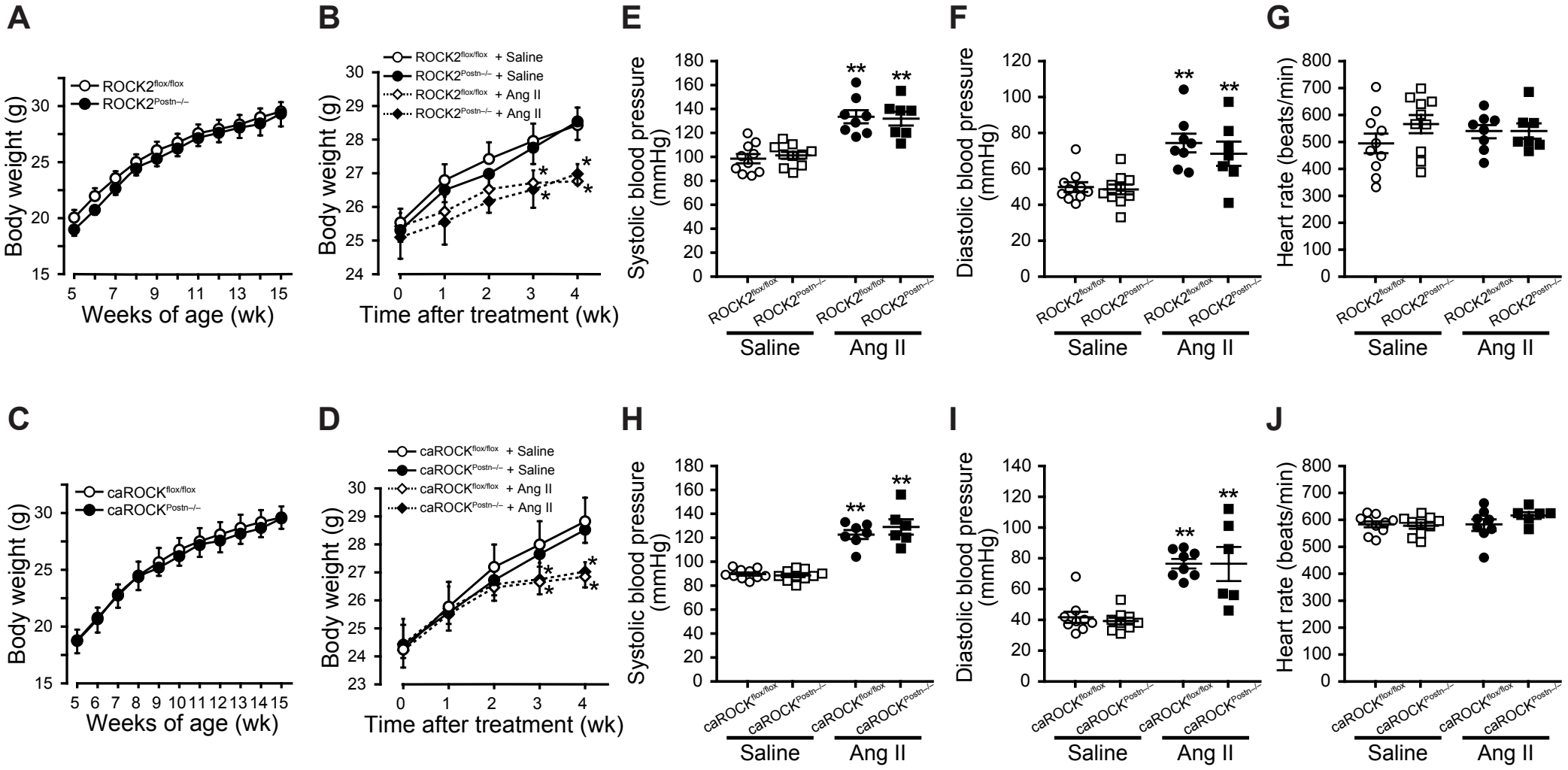
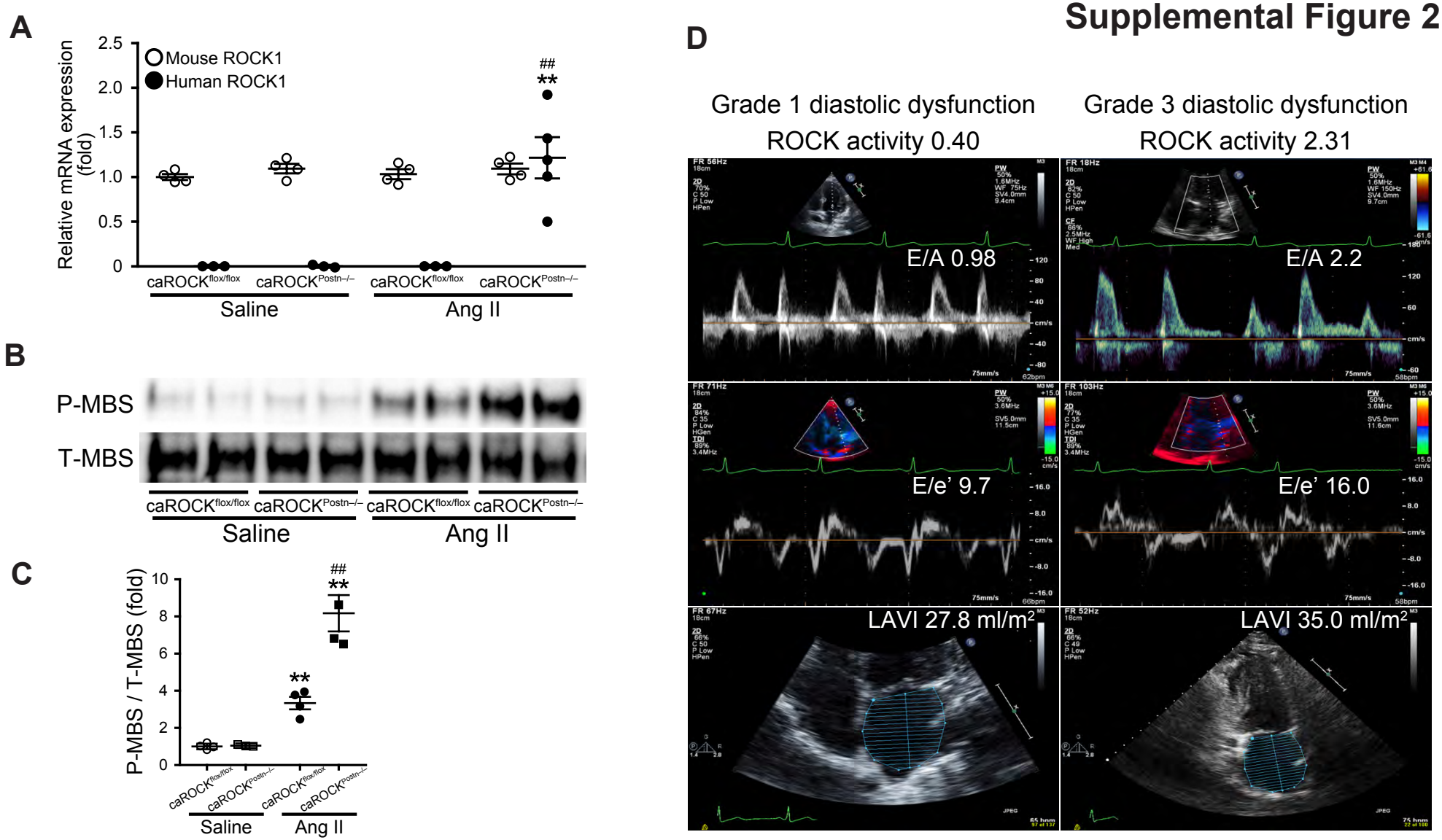
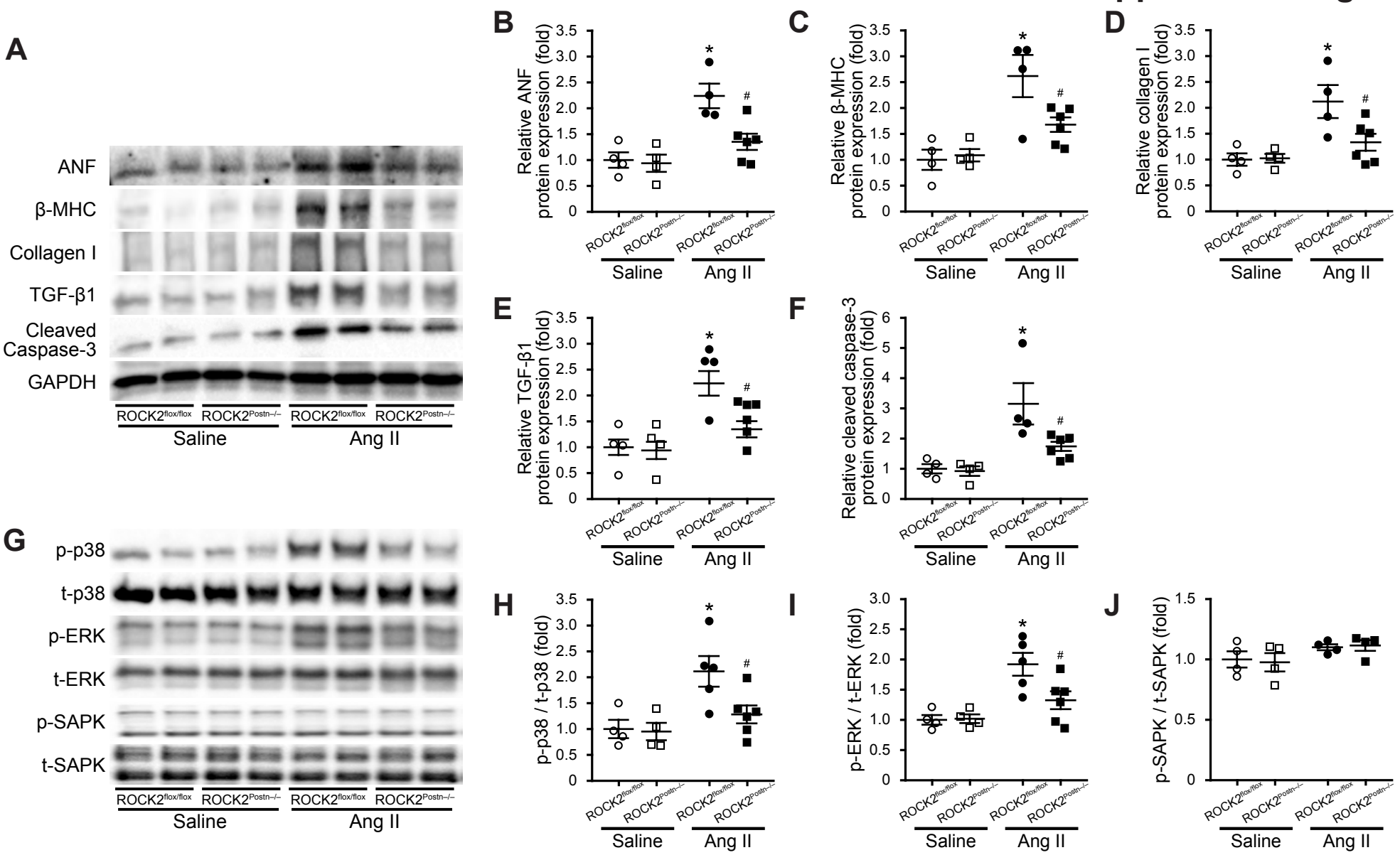


Supplemental Figure 1. Body weight and hemodynamic changes in each genotype of mice after saline or angiotensin II (Ang II) infusion. (**A**–**D**) Body weight gain over time and during infusion of saline or Ang II infusion for 4wk in fibroblast-specific ROCK2-deficient ($ROCK2^{Postn-/-}$) and littermate control ($ROCK2^{flox/flox}$) mice, and fibroblast-specific constitutively active knock-in ROCK ($caROCK^{Postn-/-}$) and littermate control ($caROCK^{flox/flox}$) mice (*n*=10 each). (**E**–**J**) Systolic and diastolic blood pressure and heart rate among the four genotypes at 4wk after saline or Ang II infusion (*n*=6-10 each). **P*<0.05, ***P*<0.01 vs. saline-treated each genotype. Data are expressed as mean±SEM. *P* values were calculated using unpaired Student's *t*-test or one-way ANOVA with Tukey's HSD test.



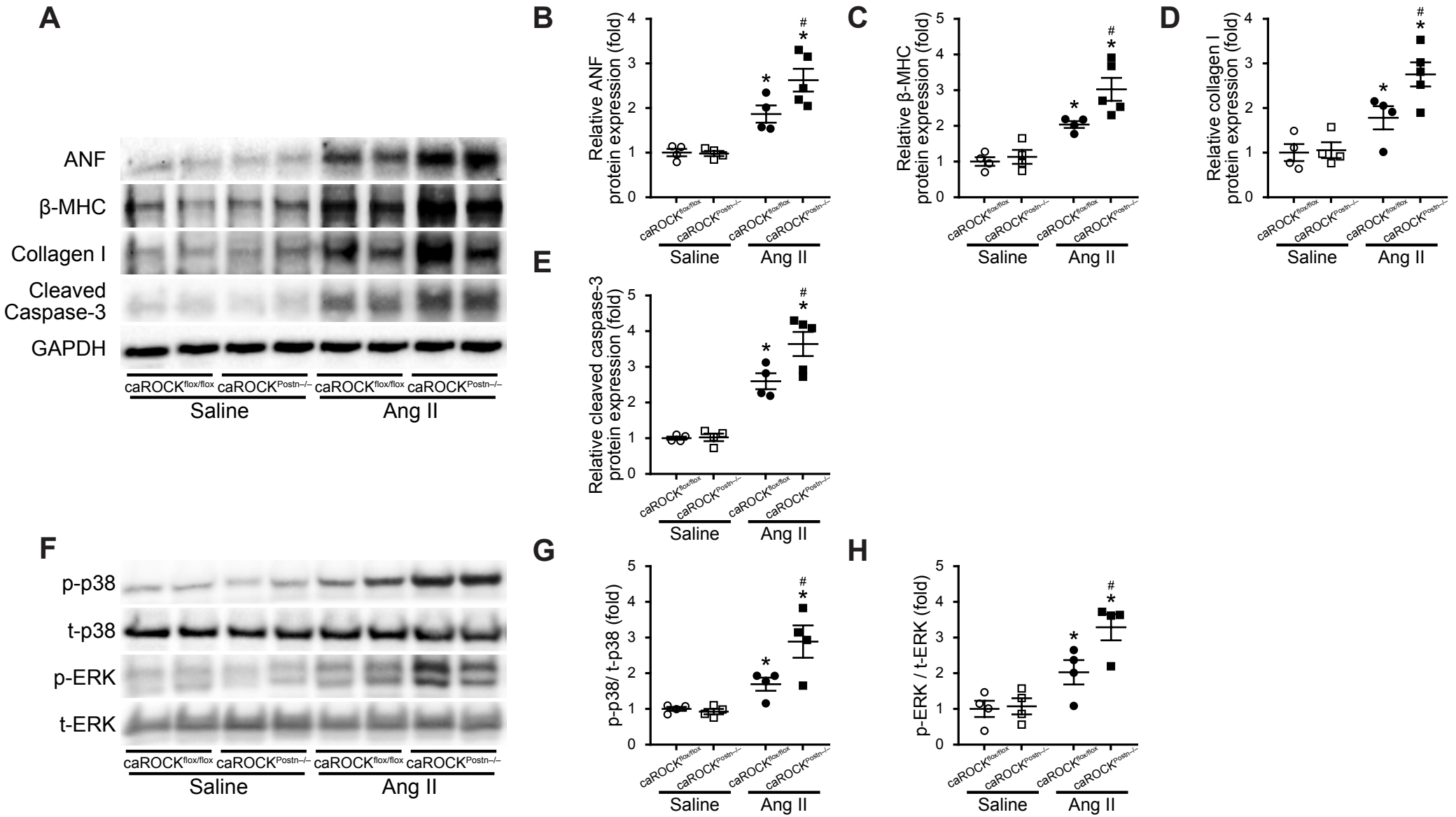
Supplemental Figure 2. Increased ROCK activity in cardiac fibroblasts (CFs) from fibroblast-specific constitutively active ROCK knock-in (caROCK$^{Postn-/-}$) mice after angiotensin II (Ang II) infusion. (**A**) Quantitative RT-PCR analysis of mouse ROCK1 and human ROCK1 mRNA expression in CFs isolated from caROCK$^{Postn-/-}$ and littermate control (caROCK$^{flox/flox}$) mice at 4wk after saline or Ang II infusion (*n*=3-4 each). $^{**}P$<0.01 vs. human ROCK1 expression in CFs from saline-treated caROCK$^{Postn-/-}$ mice. $^{\#\#}P$<0.01 vs. human ROCK1 expression in CFs from Ang II-treated caROCK$^{flox/flox}$ mice. (**B**) Representative immunoblots of ROCK activity, as assessed by the ratio of phosphorylated form of the myosin-binding subunit (MBS) to total MBS (p-MBS/t-MBS), in CFs from caROCK$^{Postn-/-}$ and caROCK$^{flox/flox}$ mice at 4wk after saline or Ang II infusion. (**C**) Quantitative analysis of ROCK activity in CFs from caROCK$^{Postn-/-}$ and caROCK$^{flox/flox}$ mice (*n*=3-4 each). $^{**}P$<0.01 vs. saline-treated each genotype. $^{\#\#}P$<0.01 vs. Ang II-treated caROCK$^{flox/flox}$ mice. Data are expressed as mean±SEM. *P* values were calculated using one-way ANOVA with Tukey' s HSD test. (**D**) Representative echocardiographic images of transmitral velocity (E/A) ratio, mitral E/e' ratio, and left atrial volume index (LAVI) from patients with grade 1 diastolic dysfunction and those with grade 3 diastolic dysfunction and higher leukocyte ROCK activity.

Supplemental Figure 3.



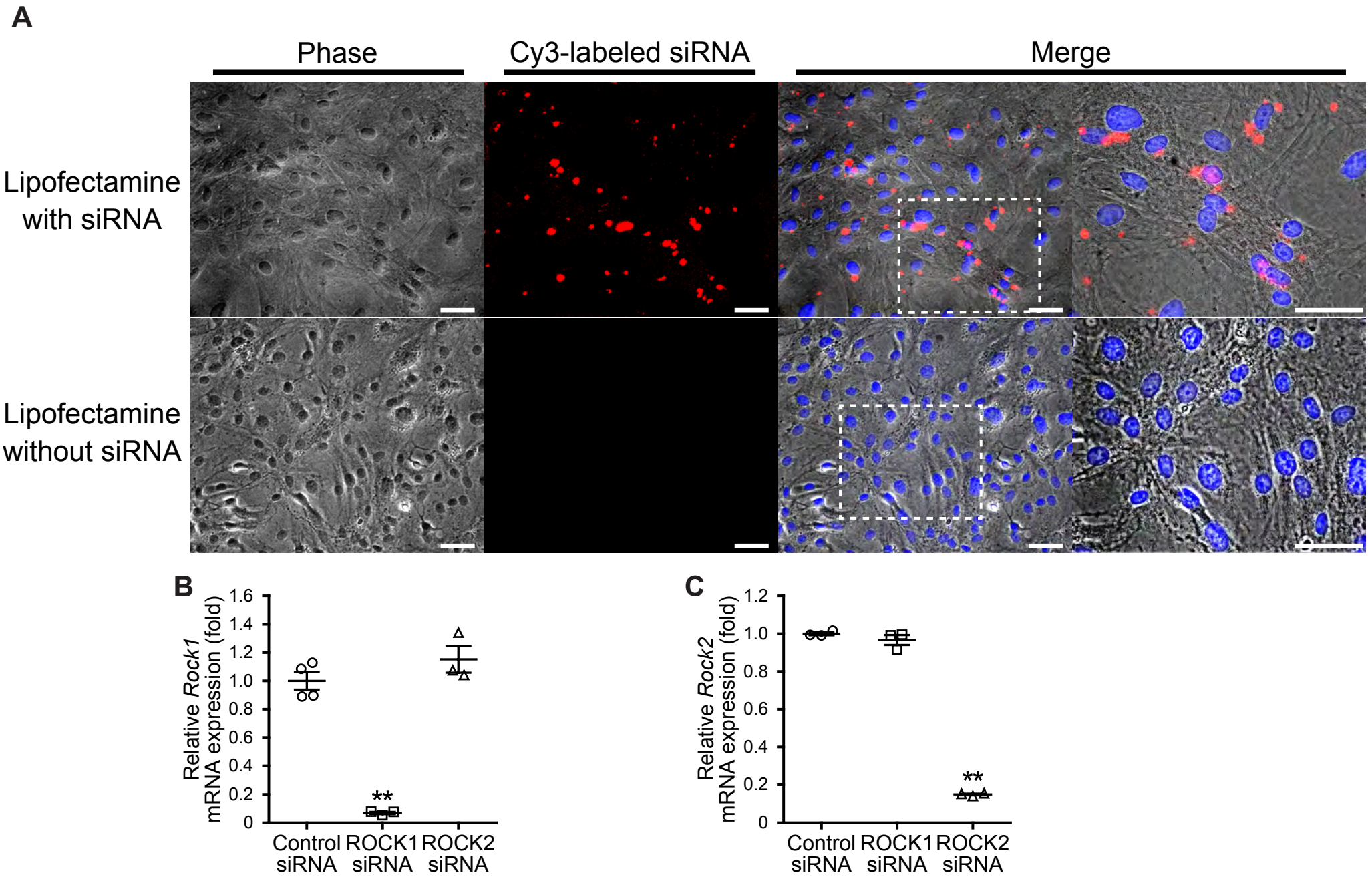
Supplemental Figure 3. Down-regulation of signaling pathways involved in cardiac remodeling and apoptosis in hearts from fibroblast-specific ROCK2-deficient (ROCK2$^{Postn-/-}$) mice treated with angiotensin II (Ang II). (**A**–**F**) Representative immunoblots and densitometric quantification of hypertrophic markers of atrial natriuretic factor (ANF) and beta-myosin heavy chain (β-MHC), a fibrotic marker of collagen type I, TGF-β1, and an apoptosis marker of cleaved caspase-3 in heart tissues from ROCK2$^{Postn-/-}$ and littermate control (ROCK2$^{flox/flox}$) mice at 4 wk after saline or Ang II infusion. (*n*=4-6 each). (**G**–**J**) Representative immunoblots of mitogen-activated protein kinases (p38 MAPK, ERK, and SAPK) and densitometric quantification of phosphorylated p38 MAPK, ERK, and SAPK, normalized to respective total protein, in heart tissues from ROCK2$^{Postn-/-}$ and ROCK2$^{flox/flox}$ mice treated with saline or Ang II (*n*=4-6 each). **P*<0.05 vs. saline-treated ROCK2$^{flox/flox}$ mice. $^{\#}$*P*<0.05 vs. Ang II-treated ROCK2$^{flox/flox}$ mice. Data are expressed as mean±SEM. *P* values were calculated using one-way ANOVA with Tukey' s HSD test.

Supplemental Figure 4.



Supplemental Figure 4. Up-regulation of signaling pathways involved in cardiac remodeling and apoptosis in hearts from fibroblast-specific constitutively active knock-in ROCK (caROCK$^{Postn-/-}$) mice treated with angiotensin II (Ang II). (**A**–**E**) Representative immunoblots and densitometric quantification of hypertrophic markers of atrial natriuretic factor (ANF) and beta-myosin heavy chain (β-MHC) and, a fibrotic marker of collagen type I, and an apoptosis marker of cleaved caspase-3 in heart tissues from caROCK$^{Postn-/-}$ and littermate control (caROCK$^{flox/flox}$) mice at 4 wk after saline or Ang II infusion (n=4-5 each). (**F**–**H**) Representative immunoblots and densitometric quantification of mitogen-activated protein kinases (p38 MAPK and ERK), normalized to respective total protein, in heart tissues from caROCK$^{Postn-/-}$ and caROCK$^{flox/flox}$ mice treated with saline or Ang II (n=4 each).*P<0.05 vs. saline-treated each genotype. $^{\#}P$<0.05 vs. Ang II-treated caROCK$^{flox/flox}$ mice. Data are expressed as mean±SEM. P values were calculated using one-way ANOVA with Tukey' s HSD test.

Supplemental Figure 5.



Supplemental Figure 5. Effective siRNA knockdown of ROCK1 and ROCK2 expression in rat neonatal cardiac fibroblasts (RNCFs). (**A**) Representative phase and fluorescence images of RNCFs transfected with or without Cy3-labeled (red) control siRNA using Lipofectamine RNAiMax Reagent (*n*=3 each). Nuclei are stained with DAPI (blue). Scale bars, 50 um. (**B** and **C**) Quantitative RT-PCR analysis of *Rock1* and *Rock2* mRNA expression in RNCFs transfected with ROCK1 siRNA or ROCK2 siRNA (*n*=3-4 each). ***P*<0.01 vs. RNCFs transfected with control siRNA. Data are expressed as mean±SEM. *P* values were calculated using one-way ANOVA with Tukey' s HSD test.