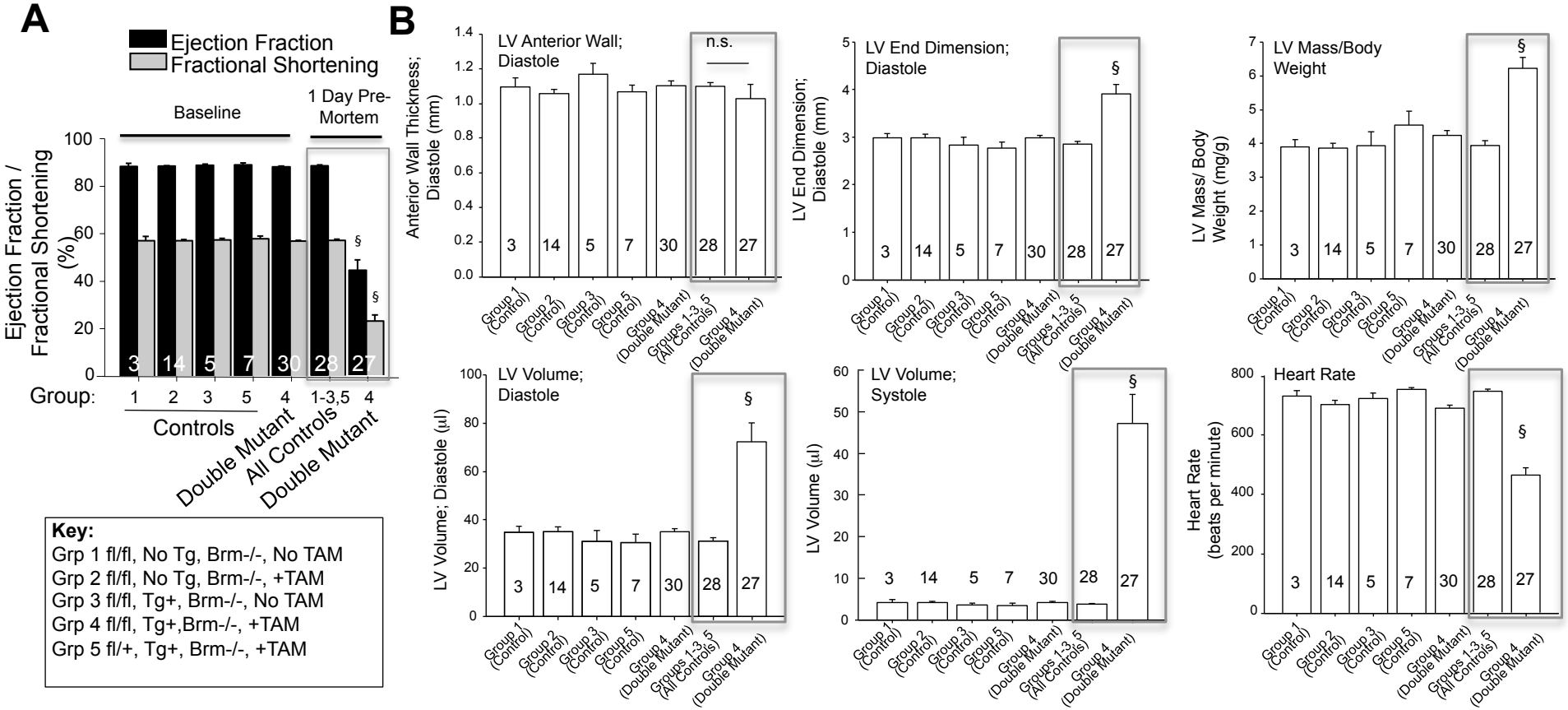

Figure 1. *Brg1/Brm* double mutants undergo arrhythmias and heart failure. (A) Echocardiogram-based measurements of ejection fraction % (black histograms) and fractional shortening % (gray histograms) in *Brg1/Brm* double mutants (Group 4) and control groups at baseline (prior to loss of *Brg1 via* tamoxifen) and at 1-day pre-mortem (histograms enclosed by gray box at right). See key below for description of each numbered control group. (B) Six panels show left-ventricle morphometrics and heart rate, as indicated, with first 5 histograms representing baseline measurements and last 2 histograms enclosed by gray box representing 1-day pre-mortem measurements. Data represent means ± SEM with the number of mice per group indicated in each histogram. One-Way Analysis of Variance was performed followed by an all pair-wise multiple comparison procedure (Holm-Sidak method) with significant differences indicated (§, $p<0.001$ vs. all other groups).

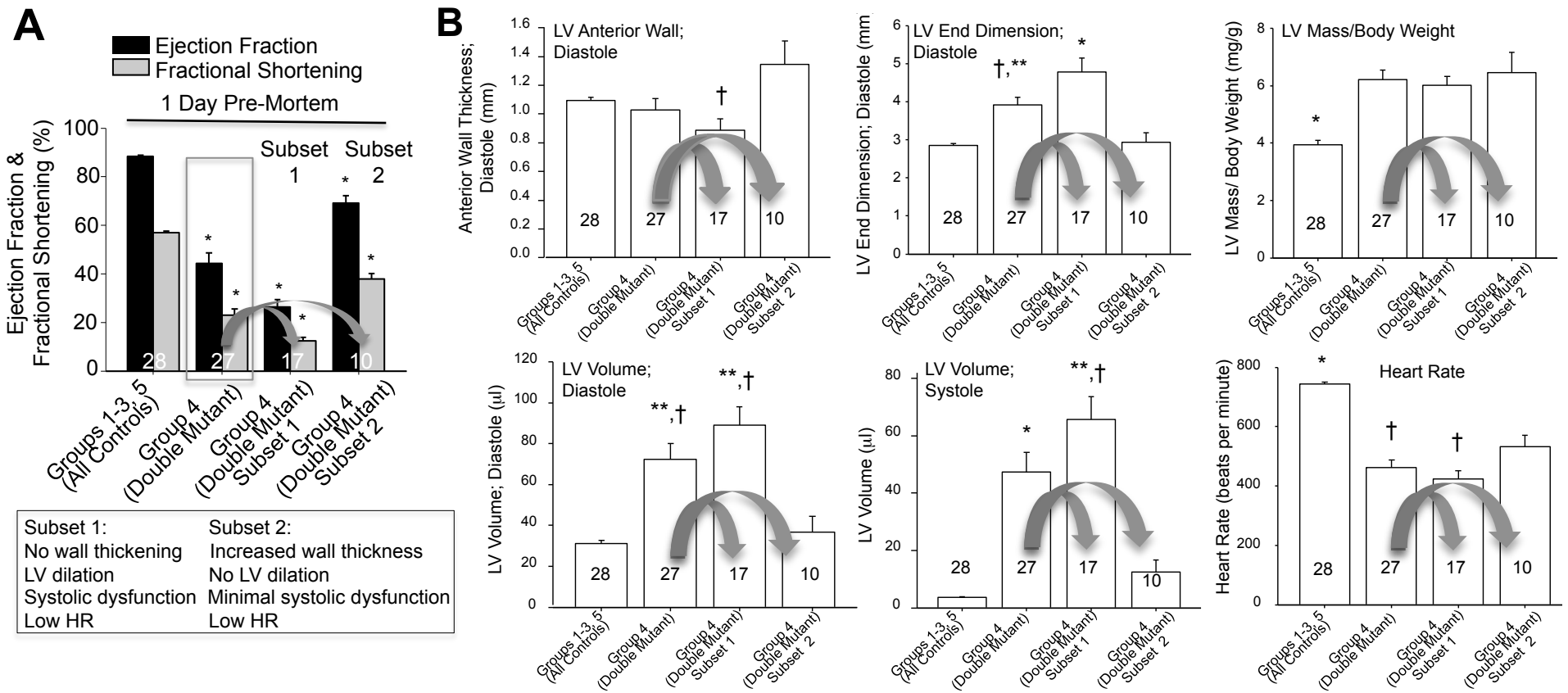

Figure 2. Two subphenotypes in *Brg1/Brm* double mutants. (A) The ejection fraction % and fractional shortening % data at 1-day pre-mortem from Fig. 1A are reproduced at the left. The double-mutant values are enclosed by a gray box. These data are juxtaposed with 1-day pre-mortem data from the double mutants separated out into two subsets (highlighted by arrows and histograms labeled Subset 1 and Subset 2) where the phenotypes differed with respect to wall thickening, LV dilation, and systolic dysfunction, but not heart rate (HR). (B) Six panels show left-ventricle morphometrics and heart rate that are the same as Fig. 1B except only 1-day pre-mortem data are shown and the double mutant data are shown combined and separated out into the two subgroups as indicated. Data represent means ± SEM with the number of mice per group indicated in each histogram. One-Way Analysis of Variance was performed followed by an all pair-wise multiple comparison procedure (Holm-Sidak method) with significant differences indicated (*p<0.001 vs. all other groups; **p<0.05 vs. Column 1; †p<0.01 vs. Column 4).

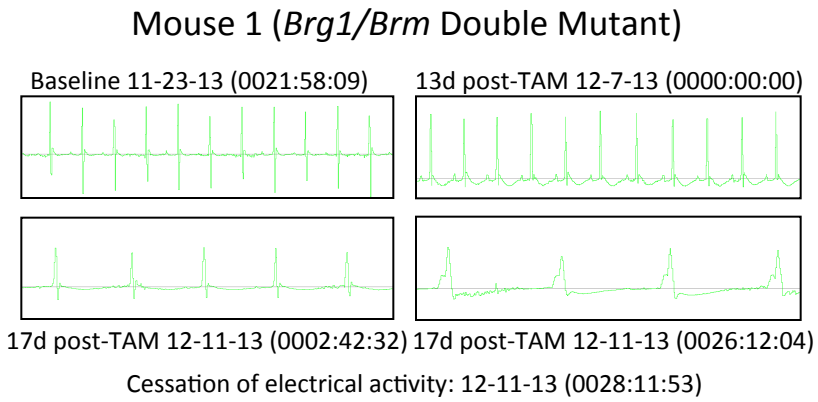

Figure 3. *Brg1/Brm* double mutants have conduction defects and die because of atrioventricular blockage. ECG data from double mutant and control mice at baseline and at 13 and 17 days following tamoxifen (TAM)-induced loss of *Brg1* that includes measurements in the hours preceding death.

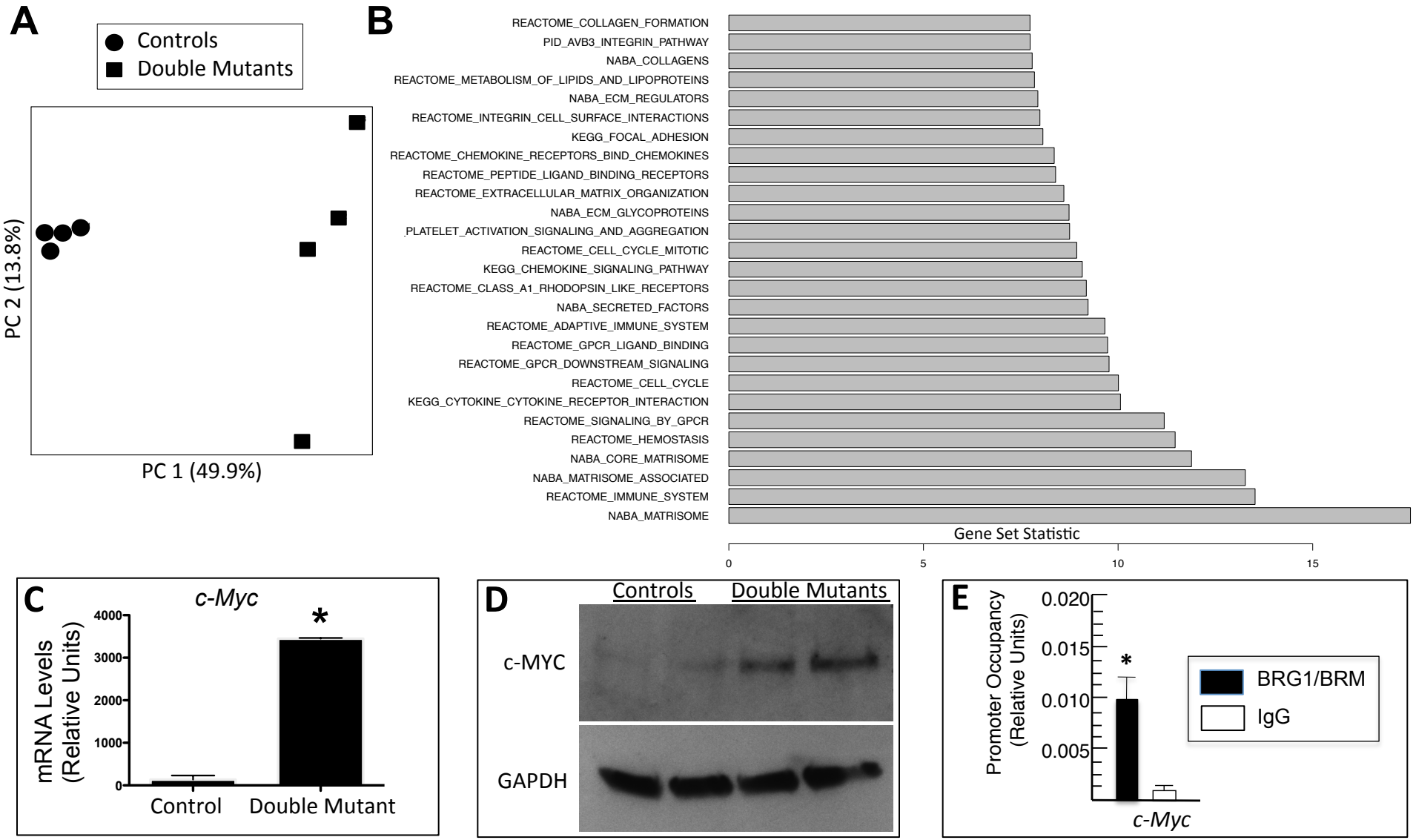

Figure 4. BRG1/BRM transcriptional targets including *c-Myc*. (A) Principal component (PC) analysis of controls and *Brg1/Brm* double mutants based on their transcriptome profiles. (B) Significantly enriched pathways among the genes differentially expressed between *Brg1/Brm* double mutants and controls. All of the listed pathways have an FDR < 0.05, and the gene set statistic on the x-axis represents the z score transformation of the mean of all genes in a set. (C) RT-qPCR analysis of *c-Myc* mRNA levels normalized to *Gapdh* in control and double-mutant hearts. Data are presented as means ± SEM based on 5 independent experiments with significant differences indicated (*p<0.05). (D) Western blot analysis of heart protein lysates from controls and double mutants probed with antibodies specific for c-MYC and GAPDH as a loading control. (E) Quantitative ChIP assays demonstrating BRG1/BRM occupancy at the *c-Myc* promoter in wild-type mouse heart. Histograms show the relative enrichment by comparing each ChIP sample to input by qPCR (means ± SEM for three independent samples, (*p<0.05).

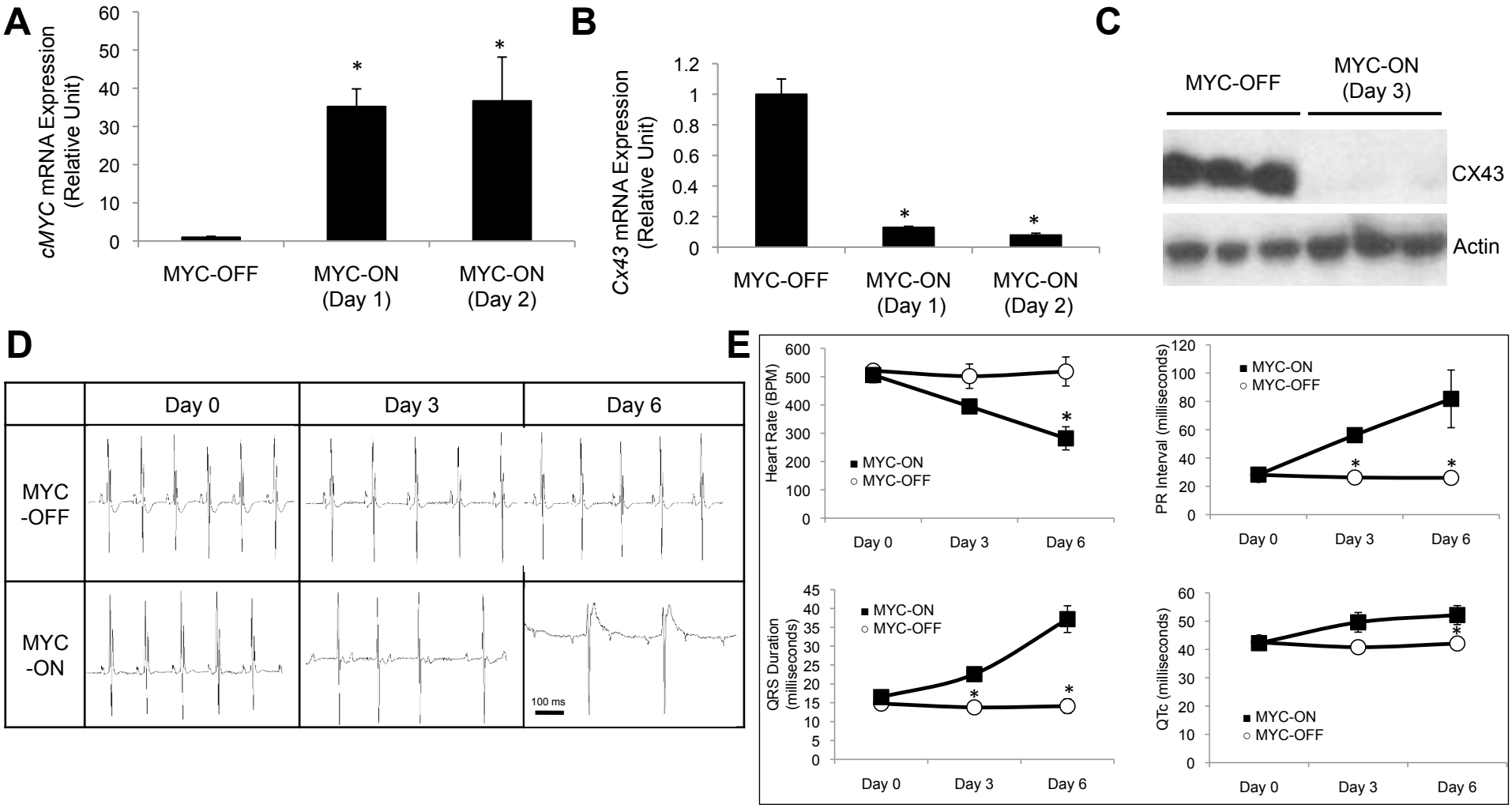

Figure 5. *c-MYC* gain-of-function in cardiomyocytes results in cardiac conduction defects that phenocopy *BRG1/BRM* loss-of-function. (A) RT-qPCR analysis of *c-MYC* mRNA levels in the heart of an inducible transgenic mouse line prior to induction (MYC-OFF) and 24-48 hours after DOX-mediated induction to overexpress *c-Myc* (MYC-ON, Day 1 and Day 2). Data are normalized to *Gapdh* and presented as means ± SEM based on 5 independent experiments with significant differences indicated (*p<0.05). (B) RT-qPCR analysis of *Cx43* mRNA levels in heart of the same transgenic mouse line. Data are normalized to *Gapdh* and presented as means ± SEM based on 5 independent experiments with significant differences indicated (*p<0.05). (C) Representative western blot of CX43 protein levels in heart of same transgenic mouse line prior to induction (MYC-OFF) and after DOX-mediated induction (MYC-ON, Day 3). Actin serves as a loading control. 3 independent samples for MYC-OFF and MYC-ON are shown. (D) ECG sample trace readings from 3 MYC-OFF controls and 4 MYC-ON mice showing Wenckebach second-degree heart block by 3 days of DOX-induced c-MYC induction and a complete heart block by day 6. (E) Four panels showing ECG-based measurements from 3 MYC-OFF controls and 4 MYC-ON mice at three time points relative to DOX-mediated induction. The plots show significant differences (*p<0.05) in heart rate, PR interval, QRS duration, and QTc.

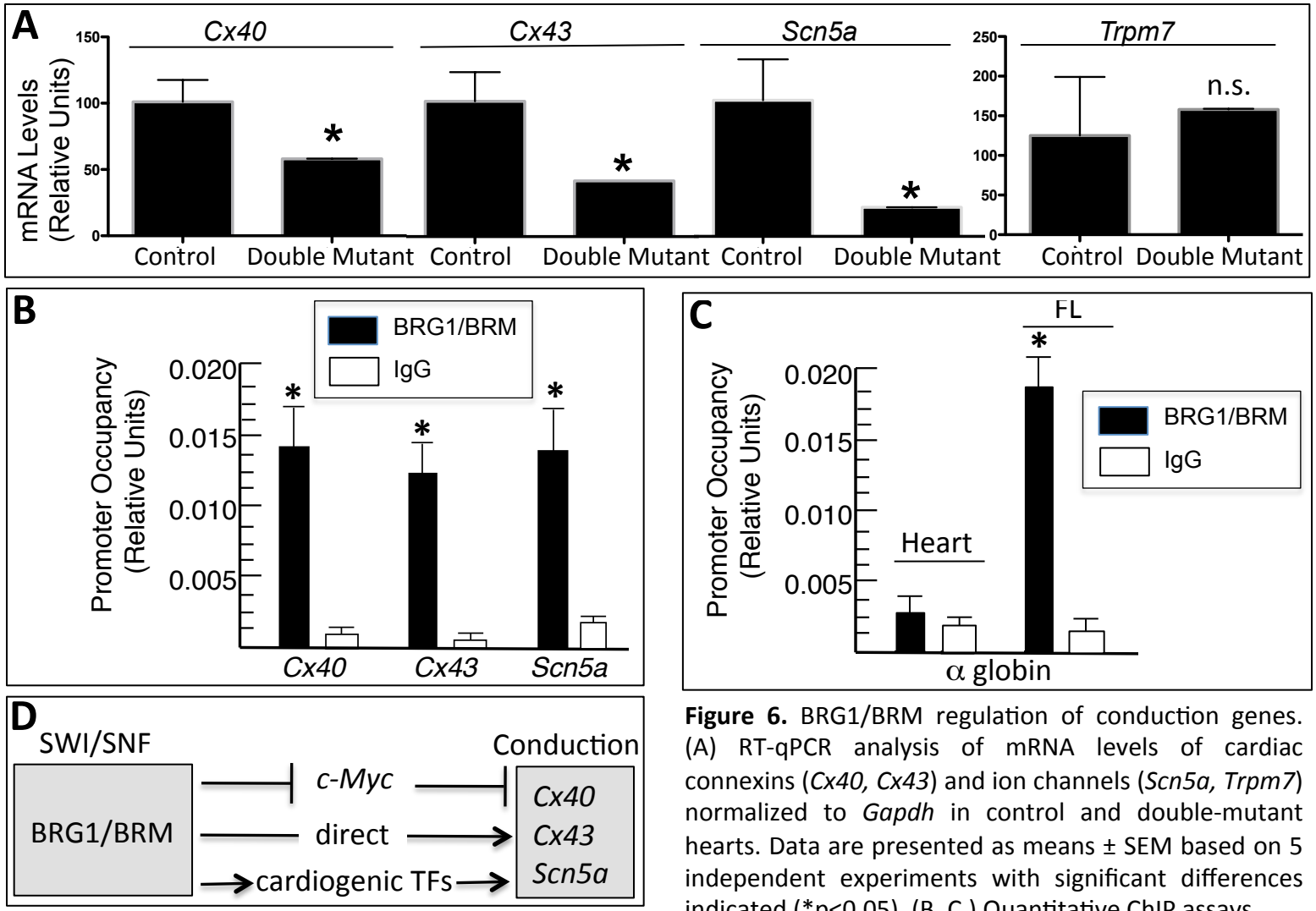

Figure 6. BRG1/BRM regulation of conduction genes. (A) RT-qPCR analysis of mRNA levels of cardiac connexins (*Cx40, Cx43*) and ion channels (*Scn5a, Trpm7*) normalized to *Gapdh* in control and double-mutant hearts. Data are presented as means ± SEM based on 5 independent experiments with significant differences indicated (*p<0.05). (B, C,) Quantitative ChIP assays demonstrating BRG1/BRM occupancy at the *Cx40, Cx43*, and *Scn5a* promoters in wild-type mouse heart tissues (B) and at the α-globin locus in wild-type heart and fetal liver (FL) tissues (C). IgG immunoprecipitations serve as a negative control. Histograms show the relative enrichment by comparing each ChIP sample to input by qPCR (means ± SEM for three independent samples, (*p<0.05). (D) Working model. The BRG1 and BRM catalytic subunits of SWI/SNF complexes directly and indirectly activate the expression of *Cx40, Cx43*, and *Scn5a* to facilitate conduction in cardiomyocytes. The direct regulation is based on ChIP assays demonstrating occupancy at each promoter. The indirect regulation is mediated by inhibition of an inhibitor (*c-Myc*) and activation of an activator (cardiogenic transcription factors *Tbx, Nkx2-5, Mef2c*).

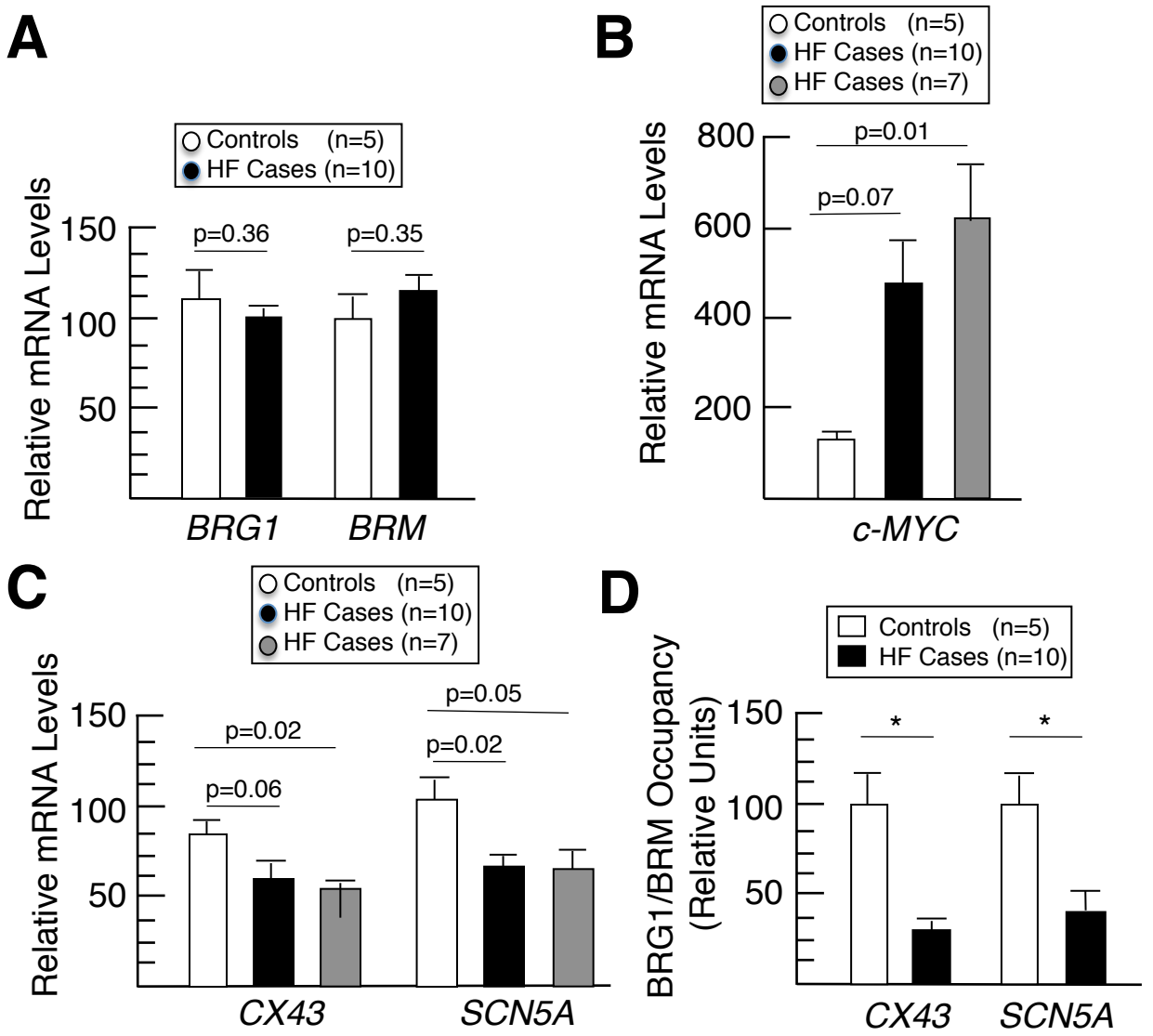

Figure 7. BRG1/BRM occupancy and expression of conduction genes is attenuated in human heart failure cases, while *c-MYC* is overexpressed. (A-C) RT-qPCR analysis of mRNA levels for human *BRG1* and *BRM* (A), *c-MYC* (B), and *CX43* and *SCN5A* (C) normalized to *GAPDH* mRNA levels. Data are presented as means ± SEM based on 5 controls (white), 10 heart failure cases (black), and a subset of heart failure cases with elevated *c-MYC* mRNA levels (gray, n=7) (as opposed to the other 3 heart failure cases that did not have elevated *c-MYC*) with p-values indicated. (D) Quantitative ChIP assays measuring BRG1/BRM occupancy at the human *CX43* and *SCN5A* promoters in cardiac tissue from 5 controls and 10 heart failure cases. Each ChIP qPCR was normalized to input, and the relative enrichments are shown as means ± SEM with significant differences indicated (*p<0.05).