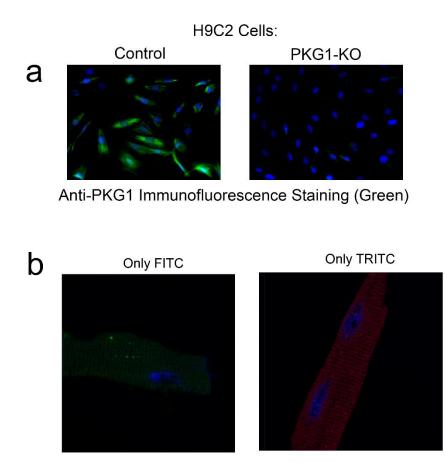
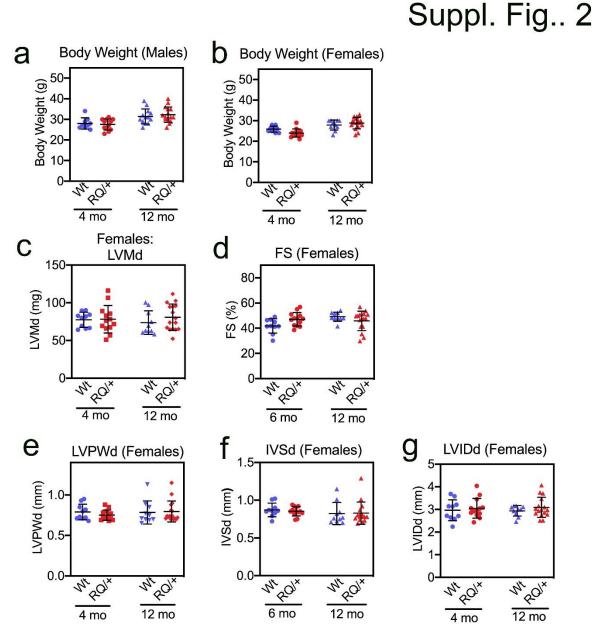
Suppl. Fig. 1



Cardiac Myocytes: Secondary Antibodies only

Supplemental Figure 1: Immunofluorescence Controls.

- (a) Validation of the PKG1 antibody: In H9C2(2-1) myoblasts from the ATCC (CRL-1446), PKG1 was deleted using CRISPR/Cas9. Wild type and PKG1-knockout cells were fixed, permeabilized and stained with the PKG1-specific antibody and a FITC-labeled secondary antibody as described in Methods. Nuclei were counterstained with Hoechst 33342.
- (b) Immunofluorescence negative controls: Cardiac myocytes from wild type mice were fixed, permeabilized, and stained as described in Methods, using only FITC-labeled or TRITC-labeled antibodies in the absence of primary antibodies, with nuclei counterstained as in panel a.



Supplemental Figure 2: Body weights in male and female mice, and basal cardiac parameters in female wild type and Prkg1^{RQ/+} mice.

(a,b) Bodyweights of male and female wild type (WT) and $Prkg1^{RQ/+}$ (RQ/+) mice, measured at 4 and 12 months of age.

(c-g) Cardiac parameters measured by echocardiography in 4-month and 12-month-old female WT and RQ/+ mice: (c) Left ventricular mass (LVMd) in diastole; (d) LV fractional shortening (FS); (e) LV posterior wall thickness in diastole (LVPWd); (f) inter-ventricular septum thickness in diastole (IVSd); (g) LV inner diameter in diastole (LVIDd).