Redox Control of Protein Kinase G I alpha Fine-Tunes the Frank-Starling Law of the Heart in Vivo by Regulating Diastolic Relaxation

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The Frank-Starling law describes a regulatory mechanism that allows the amount of blood that enters the heart to be precisely matched with the amount that is pumped out to the arterial circulation, independently of external regulation. Mechanisms that contribute to the Frank-Starling response include stretch-induced increases in myofilament overlap, myofilament Ca2+ sensitivity and actin-myosin cross-bridge formation. Recently, it was reported that there is also an increased production of oxidants during diastolic stretch, which are involved in regulation of cardiac Ca2+ cycling and contractile performance. Here we show that myocardial stretch mediates oxidation of cysteine 42 in protein kinase G Iα (PKGIα), causing it to form a disulfide dimer which is catalytically active. An unbiased phosphoproteomic study revealed that this disulfide-activated PKGIα selectively phosphorylates phospholamban (PLN) Ser16. Isothermal titration calorimetry data suggest that the interaction between PLN and the reduced form of PKGIα is markedly weaker than when the kinase is oxidised to the disulfide. Hearts isolated from C42S PKGIα knock-in (KI) mice, in which the kinase is resistant to oxidant-induced activation, were deficient in PLN Ser16 phosphorylation and displayed impaired end-diastolic relaxation and Frank-Starling responses compared to wild-type (WT). Ventricular myocytes isolated from KI mice were significantly deficient in their systolic Ca2+ transient and SR Ca2+ content and had ~50 % slower rate of Ca2+ decay compared to WT cells. In vivo analysis of cardiac function by echocardiography and using pressure-volume catheter showed KI hearts had a slower diastolic relaxation leading to an elevated end-diastolic pressure and an impaired Frank-Starling mechanism, in agreement with isolated heart data. Furthermore, in the absence of this redox control mechanism as is the case in the KI, the peak force the heart generated in vivo from beat-to-beat was erratic. We conclude that myocardial stretch-induced reactive oxygen species (ROS) production causes oxidative activation of PKGIα, fine-tuning the Frank-Starling response.