Differential role of *S*-nitrosylation and the NO–cGMP–PKG pathway in cardiac contractility

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Abstract

The role of nitric oxide (NO) in cardiac contractility is complex and controversial. Several NO donors have been reported to cause positive or negative inotropism. NO can bind to guanylate cyclase, increasing cGMP production and activating PKG. NO may also directly *S*-nitrosylate cysteineresidues of specific proteins. We used the isolated rat heart preparation to test the hypothesis that the differential inotropic effects depend on the degree of NO production and the signaling recruited. SNAP (*S*-nitroso-*N*-acetylpenicillamine), a NO donor, increased contractility at 0.1, 1 and 10 μM. This effect was independent of phospholamban phosphorylation, was not affected by PKA inhibition with H-89 (*N*-[2((*p*-bromocinnamyl)amino)ethyl]-5-isoquinolinesulfonamide), but it was abolished by the radical scavenger Tempol (4-hydroxy-[2,2,4,4]-tetramethyl-piperidine-1-oxyl). However, at 100 μM SNAP reduced contractility, effect reversed to positive inotropism by guanylyl cyclase blockade with ODQ (1*H*-[1,2,4]oxadiazolo[4,3-*a*]quinoxalin-1-one), and abolished by PKG inhibition with KT5823, but not affected by Tempol. SNAP increased tissue cGMP at 100 μM, but not at lower concentrations. Consistently, a cGMP analog also reduced cardiac contractility. Finally, SNAP at 1 μM increased the level of *S*-nitrosylation of various cardiac proteins, including the ryanodine receptor. This study demonstrates the biphasic role for NO in cardiac contractility in a given preparation; furthermore, the differential effect is clearly ascribed to the signaling pathways involved. We conclude that although NO is highly diffusible, its output determines the fate of the messenger: low NO concentrations activate redox processes (*S*-nitrosylation), increasing contractility; while the cGMP–PKG pathway is activated at high NO concentrations, reducing contractility.