

## **Effects of a Human Inhibitor-1 Polymorphism (G147D) on Calcium Homeostasis and Contractility In Adult Rat Cardiomyocytes**

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The sarcoplasmic reticulum (SR) serves as a source and sink for  $\text{Ca}^{2+}$  during contraction and relaxation of the heart, respectively. Disruption of this process plays a critical role in inducing heart failure. Phosphorylation of the ryanodine receptor (RyR), a  $\text{Ca}^{2+}$  release channel of the SR, by protein kinase A (PKA) is believed to be integral to calcium cycling within the cardiomyocyte. Protein Phosphatase-1 (PP1) reverses the effects of PKA through dephosphorylation of RyR. However, PP1 is inhibited by endogenous Inhibitor-1 (I-1). The aim is to determine whether a human polymorphism of I-1, G147D, which has been known to disturb Ca-cycling and contractility, affects phosphorylation of RyR. Our hypothesis is that this specific polymorphism interferes with cardiac function through the altered phosphorylation of RyR. Isolated myocytes from adult rat hearts were infected with adenoviruses containing the G147D mutant I-1, or wild type I-1, or empty vector. Subsequently, basal and phosphorylated states of RyRs were determined by quantitative western blotting. Preliminary results showed that in the presence of isoproterenol stimulation, which activates the PKA pathway, the phosphorylation of RyR was decreased in the G147D mutant as compared to wild type I-1. This indicates that the mutant I-1 may affect cardiomyocyte calcium cycling via reduced phosphorylation of RyR, resulting in altered heart function.