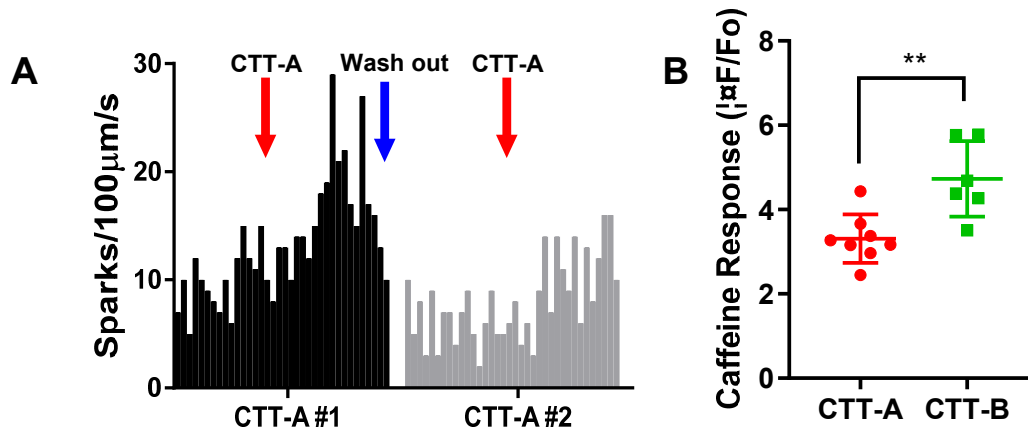
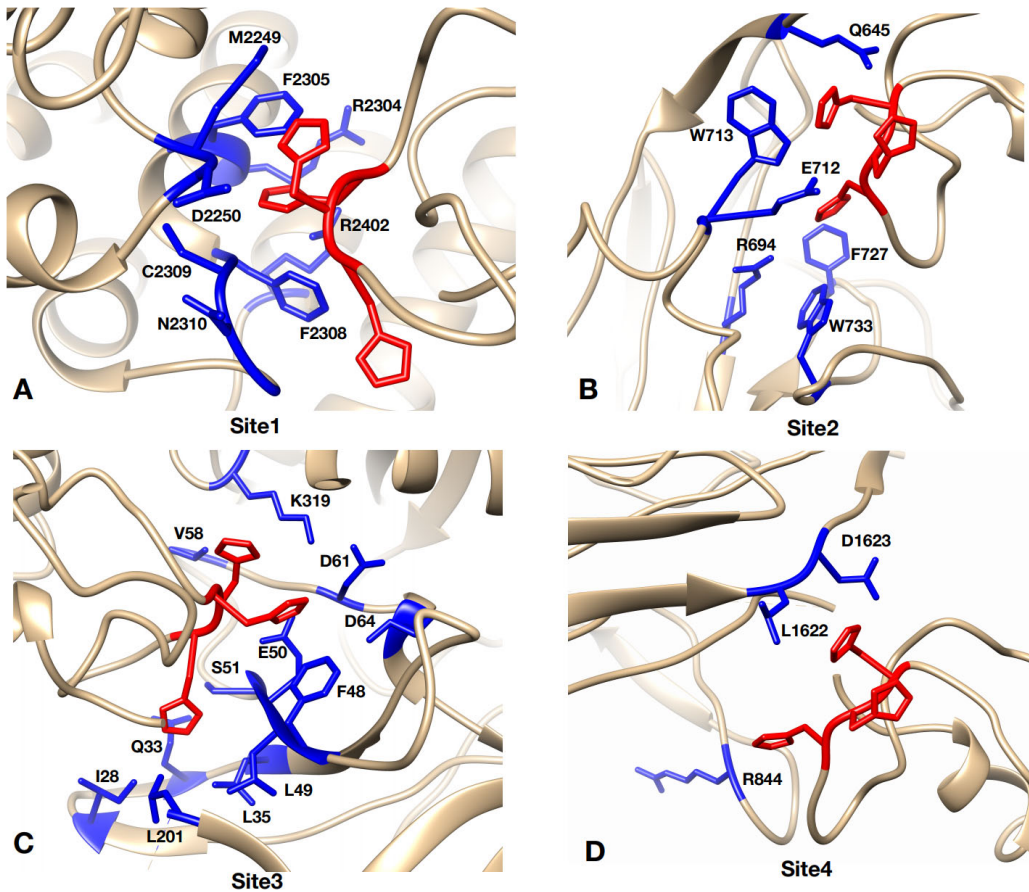


# Supplemental Material



## Online Figure I. Effect of CTT-A on $\text{Ca}^{2+}$ sparks is reversible.

A. First addition of CTT-A ( $10 \mu\text{M}$ ) led to transient stimulation of  $\text{Ca}^{2+}$  spark activity in permeabilized TRIC-A<sup>-/-</sup> cardiomyocytes. Wash out of CTT-A returned the  $\text{Ca}^{2+}$  spark activity to the basal level. Re-addition of CTT-A resulted in reactivation of  $\text{Ca}^{2+}$  sparks B. Reduced Caffeine response in cardiomyocytes with the presence of CTT-A compared to CTT-B. ( $P=0.0034$ , unpaired t-test)



**Online Figure II. Zoom-in picture of CCT-A interacting with closed state of hRyR<sub>2</sub>.**

The backbones of CCT-A and hRyR<sub>2</sub> are plotted in ribbon representation and colored grey. The histidine-rich motif is plotted in stick mode and colored red. The residues of hRyR<sub>2</sub> interacting with CCT-A are drawn in stick mode, labeled, and colored blue. A-D represents the four sites of CCT-A interaction with hRyR<sub>2</sub>

**Online Movie I and II.** Continuous x-y images of spontaneous Ca<sup>2+</sup> waves were acquired using the resonant scan mode of the Nikon A1R confocal microscope (at a rate of 130 ms per frame). TRIC-A<sup>-/-</sup> cardiomyocytes (**II**) showed a slower Ca<sup>2+</sup> wave propagation speed compared to WT cardiomyocytes (**I**) in 10 mM extracellular Ca<sup>2+</sup> condition.

**Online Movie III and IV.** Real-time movies of store-overload induced Ca<sup>2+</sup> waves in WT and TRIC-A<sup>-/-</sup> cardiomyocytes. The recording conditions were the same as **Movie I**. In addition to the apparent slower speed of Ca<sup>2+</sup> wave propagation, fewer numbers of TRIC-A<sup>-/-</sup> cardiomyocytes (**IV**) show spontaneous Ca<sup>2+</sup> waves at 10 mM extracellular Ca<sup>2+</sup> condition compared to the WT cardiomyocytes (**III**).