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Regulation of Ryanodine Receptor Ion Channels Through Posttranslational Modifications

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I. OVERVIEW

The ryanodine receptors (RyRs) are cation-selective channels that release Ca^{2+} from an intracellular Ca^{2+} storing compartment, the endo/sarcoplasmic reticulum, during an action potential in a process known as excitation–contraction coupling. The released Ca^{2+} ions regulate a wide variety of biological functions. In striated muscle, the release of Ca^{2+} from the sarcoplasmic reticulum into the cytoplasm leads to muscle contraction. This chapter focuses on the regulation of the skeletal muscle and cardiac muscle RyRs by protein kinases and redox active species. The RyRs are 2200 kDa Ca^{2+}-gated ion channels that are regulated, in addition to Ca^{2+} and endogenous effectors such as Mg^{2+} and ATP, by cAMP-dependent protein kinase A, calmodulin-dependent kinase II, protein kinase C, and protein phosphatases 1 and 2A. Thiols that serve as targets for reactive oxygen and nitrogen molecules determine the redox state and modulate the activity of the RyRs.

II. INTRODUCTION

The RyRs are large multiprotein complexes composed of four 560-kDa RyR subunits, four small FK506 binding protein (FKBP, also known as calstabin) subunits, and multiple associated proteins that include triadin, junctin, junctophilin, protein kinases and phosphatases, and calmodulin (Fill & Copello, 2002; Franzini-Armstrong & Protasi, 1997; Meissner, 1994; Zalk, Lehnart, & Marks, 2007). There are three mammalian RyR isoforms. RyR1 is found in the sarcoplasmic reticulum (SR) membrane of skeletal muscle, while RyR2 is present in heart muscle. In mammalian cells, RyR3 is coexpressed with one or two of the other RyR isoforms at low levels. All three isoforms are present in smooth muscle (Neylon, Richards, Larsen, Agrotis, & Bobik, 1995) and brain (Furuichi et al., 1994).

The mechanisms of RyR ion channel regulation have been most extensively studied in striated muscle. RyR1 and RyR2 reside in the junctional SR membrane near plasmalemmal L-type Ca^{2+} channels. An action potential in skeletal muscle initiates L-type Ca^{2+} channel protein conformational changes that alter the conformation of RyR1 by a direct physical interaction. In cardiac muscle, an action potential results in the influx of extracellular Ca^{2+}. Both mechanisms lead to the release of Ca^{2+} from the SR and subsequent muscle contraction (Fabiato, 1983; Rios & Pizarro, 1991). Sequestration of released Ca^{2+} by the SR Ca^{2+}-transporting ATPase (SERCA) and extrusion by the Na^{+}–Ca^{2+} exchanger restore the myofibrillar Ca^{2+} concentration from 10^{-6}–10^{-5} to ~10^{-7} M, causing muscle to relax.

The RyRs share ~70% sequence homology, with the greatest homology in the carboxyl-terminal region. In all isoforms, the C-terminal portion of the protein contains the transmembrane domain. Hydropathy analysis suggests between 4 and 12 transmembrane segments per RyR subunit (Takeshima et al., 1989; Zorzato et al., 1990). More recent studies using green fluorescence protein inserts and protease digestion indicate that each RyR1 subunit contains six to eight transmembrane helices (Du et al., 2004; Du, Sandhu,

Khanna, Guo, & MacLennan, 2002). That six membrane spanning segments are sufficient to form a channel is supported by single channel recordings of tryptic fragments (Callaway et al., 1994), the presence of six transmembrane segments in the related inositol 1,4,5-trisphosphate receptor (Michikawa et al., 1994), and RyR cryo-electron microscopy data (Samso, Feng, Pessah, & Allen, 2009). The remaining RyR amino acids form the large catalytic cytoplasmic ‘‘foot’’ structure (Franzini-Armstrong & Protasi, 1997). As shall be discussed later, experimental evidence for several phosphorylation sites and redox-sensitive sites in the cytoplasmic structure has been described.

III. RYR1 AND RYR2 PHOSPHORYLATION

A. RyR Phosphorylation Sites

Primary sequence analysis suggests the presence of many phosphorylation sites in the large cytoplasmic foot region of the RyRs (Takeshima et al., 1989; Zorzato et al., 1990). Kinases and phosphatases that are part of the RyR1 and RyR2 multiprotein complexes include protein kinase A (PKA), Ca^{2+}/calmodulin-dependent protein kinase II (CaMKII), and protein phosphatase 1 (PP1) (Currie, Loughrey, Craig, & Smith, 2004; Dulhunty et al., 2001; Hohenegger & Suko, 1993; Marx et al., 2000; Marx, Reiken, et al., 2001; Ruehr et al., 2003). The RyR2 complex also contains protein phosphatase 2A (PP2A) (Marx et al., 2000). Protein phosphatase 2B (calcineurin) associates with RyR1 (Shin et al., 2002). Anchoring proteins mediate the interaction between RyRs and associated kinases and phosphatases through conserved leucine/isoleucine motifs. A-kinase anchoring protein targets PKA and phosphodiesterase 4D3, spinophilin targets PP1, and PR130 mediates the interaction of PP2A with RyR2 (Zalk et al., 2007).

Initial studies indicated that the RyRs have one phosphorylation site, RyR2-Ser2809 (Witcher, Kovacs, Schulman, Cefali, & Jones, 1991). Different functional effects following CaMK-, PKA-, and cGMP-dependent protein kinase phosphorylation indicated the presence of additional phosphorylation sites (Hain, Onoue, Mayrleitner, Fleischer, & Schindler, 1995; Takasago, Imagawa, Furukawa, Ogurusu, & Shigekawa, 1991). Xiao et al. (2005) observed that substitution of RyR2-Ser2809 with an alanine did not abolish PKA phosphorylation of RyR2 and by peptide mapping identified Ser2030 as a second phosphorylation site of RyR2. Wehrens, Lehnart, Reiken, and Marks (2004) showed that CaMKII uniquely phosphorylates Ser2815 near Ser2809 in recombinant RyR2 expressed in human embryonic kidney 293 cells. However, incorporation of more than one ^{32}P per monomer into the native, immunoprecipitated receptor indicated the presence of an additional CaMKII site in RyR2, in partial agreement with Rodriguez, Bhogal, and Colyer (2003) that there are four CaMKII phosphorylation sites relative to one PKA site, or eight sites based on two PKA sites per RyR2 monomer. The ryanodine receptor-calcium release channel in skeletal muscle was phosphorylated at Ser2843 (corresponding to Ser2809 in RyR2) by cAMP-, cGMP-, and CaM-dependent protein kinases (Suko et al., 1993). However, the presence of additional phosphorylation sites seems likely, since CaMKII also phosphorylated threonine residue(s).

B. RyR Modulation by PKA-Mediated Phosphorylation and the Role of FK506 Binding Proteins

During exercise and stress, β-adrenergic receptor stimulation results in increased PKA activity and changes in the activity of Ca^{2+} handling proteins involved in the release of Ca^{2+} (L-type Ca^{2+} channel, RyR2) and removal of Ca^{2+} (SERCA2a and its regulatory protein phospholamban) (Bers, 2002; Meissner, 2002). On the other hand, chronic stimulation of the sympathetic system can lead to maladaptive changes of β-adrenergic receptor signaling, cardiomyocyte death, and heart failure (Koch, 2004). *In vitro* phosphorylation of Ser2809 by CaMK, and to a lesser extent by PKA, activated the RyR ion channel isolated from cardiac

muscle (Witcher et al., 1991). PKA regulated the ATP and Mg^{2+} sensitivity of RyR2 (Hain et al., 1995; Uehara, Yasukochi, Mejia-Alvarez, Fill, & Imanaga, 2002), changed channel kinetics, and increased the sensitivty to Ca^{2+} (Valdivia, Kaplan, Ellis-Davies, & Lederer, 1995). Single channel activity was lowest when RyR2-Ser2809 was phosphorylated to about 75% (Carter, Coyler, & Sitsapesan, 2006). Either a decreased or enhanced phosphorylation increased single channel activity. Marx et al. (2000) showed that PKA phosphorylation releases the small subunit FKBP12.6 (calstabin 2) from RyR2, which increased channel sensitivity to Ca^{2+} and induced the formation of channel substates. In failing hearts, RyR2 was PKA hyperphosphorylated, depleted of the FKBP12.6 subunit, and exhibited increased channel activity. Similarly, in skeletal muscle of animal models with heart failure and patients with heart diseases, exercise was linked to hyperphosphorylation and FKBP12 depletion of the skeletal muscle RyR1, increased RyR1 channel activity, and decreased exercise capacity (Reiken, Lacampagne, et al., 2003; Ward et al., 2003). These findings led Marks and coworkers to propose that PKA hyperphosphorylation and dissociation of the small FKBP subunit from the RyRs result in increased RyR sensitivity to cytosolic Ca^{2+}, referred to as leaky SR Ca^{2+} channels, and in the case of SR, Ca^{2+} handling referred to as SR Ca^{2+} leak.

The small 12 and 12.6 kDa FK506 binding proteins are predominantly associated with RyR1 and RyR2, respectively (Lam et al., 1995). They belong to the family of immunophilins, exhibit *cis/trans* isomerase activity, and their removal is reported to functionally uncouple groups of channels and increase channel activity (Brillantes et al., 1994; Marx, Gaburjakova, et al., 2001). Mechanisms implicated in RyR2 hyperphosphorylation and FKBP depletion include increased PKA activity and loss of PP1, PPA2, and phosphodiesterase 4D from the RyR2 macromolecular complex in failing hearts (Lehnart et al., 2005; Reiken, Gaburjakova, et al., 2003; Reiken, Lacampagne, et al., 2003). PKA-mediated hyperphosphorylation of RyR1 differed from that of RyR2 in that it failed to dissociate PP1 (Reiken, Lacampagne, et al., 2003).

Chen and colleagues (Jones et al., 2008; Meng et al., 2007) showed that the Ser2030 and Ser2809 phosphorylation sites in RyR2 are not located close to the FKBP12.6 binding site, as determined by cryo-electron microscopy. Therefore, it is unlikely that PKA-mediated phosphorylation of serines directly inhibits binding of FKBP12.6 to RyR2.

In attempts to define the specificity and functional consequences of RyR1 and RyR2 phosphorylation,mutantswere prepared to mimic the dephosphorylated (alanine) or phosphorylated (aspartic acid) forms. A Ca^{2+}-dependent increase in open channel probability (P_o) was observed with recombinant RyR2-S2809D, whereas RyR2-S2815A open channel probability was comparable to WT-RyR2 (Wehrens, Lehnart, et al., 2004). The recombinant RyR1-S2843A mutant exhibited a single channel behavior similar to WT-RyR1, whereas the RyR1-S2843D mutation increased channel activity (Reiken, Lacampagne, et al., 2003). There was an increase in number of channel events, and current histograms indicated the appearance of substates similar to those observed in native, PKA-phosphorylated RyRs. In binding assays, RyR2-S2809A bound FKBP12.6 with an affinity comparable to nonphosphorylated WT-RyR2,whereas Ser2809Ddisplayed a reduced binding affinity comparable to PKA-phosphorylated WT-RyR2 (Wehrens et al., 2004). These findings suggested that RyR phosphorylation and release of the accessory FKBPs lead to dysfunctional SR Ca^{2+} handling in striated muscle.

Several laboratories have challenged the hyperphosphorylation/FKBP depletion hypothesis of Marks and colleagues, which postulates that PKA hyperphosphorylation leads to FKBP depletion and leaky RyRs. Stange, Xu, Balshaw, Yamaguchi, and Meissner (2003) expressed RyR1-Ser2843A, RyR1-Ser2843D, RyR2-Ser2809A, and RyR2-Ser2809D

phosphorylation mutants in HEK293 cells. In single channel and [^{3}H]-ryanodine binding assays, neither the skeletal nor cardiac mutants showed differences from wild type (WT) with regard to regulation by Ca^{2+}, Mg^{2+}, or ATP. Furthermore, no significant alterations in the FKBP/RyR binding ratios were observed. The PKA hyperphosphorylation/FKBP depletion hypothesis has also been disputed by Chen and colleagues (Xiao et al., 2006). FKBP12.6 bound to both the phosphorylated and nonphosphorylated forms of Ser2809. Ser-2030, but not Ser-2809, was the major RyR2 phosphorylation site responding to protein kinase A activation upon β-adrenergic stimulation in normal and failing hearts. RyR2-S2830D mutation sensitized the recombinant channel to luminal Ca^{2+}, referred to as store-overload-induced Ca^{2+} release (SOICR) (Xiao, Tian, Xie, et al., 2007).

Yano et al. (2000) assessed the functional interaction of FKBP12.6 with RyR2 in a canine model of pacing-induced heart failure, using the FKBP-interacting drug [^{3}H]-dihydro-FK506. The stoichiometry of FKBP12.6 to RyR2 was lowered from 3.6 in normal hearts to 1.6 in failing hearts. Xin et al. (2002) made the interesting observation that knockout of the FKBP12.6 gene resulted in cardiac hypertrophy in male but not female mice. Treatment with tamoxifen, an estrogen receptor α antagonist, resulted in cardiac hypertrophy, which suggested that estrogen protects the female FKBP12.6 null mice from cardiac hypertrophy. Additional studies indicated that FKBP12.6$^{-/-}$ mice exhibited exercise-induced arrhythmias that were similar to those observed in patients carrying RyR2 mutations associated with catecholaminergic polymorphic ventricular tachycardia (CPVT) (Wehrens et al., 2003). CPVT mutations decreased the binding affinity of FKBP12.6 and showed an increased single channel activity compared to WT following PKA phosphorylation. It was also shown that substitution of an aspartic residue with serine (FKBP12.6-D37S) increased the binding affinity of the FKBP12.6 mutant to PKA-phosphorylated WT-RyR2, RyR2-S2809D mutant, and CPVT-associated RyR2 mutants. Binding of the FKBP12.6 mutant restored normal channel function. Consistent with these findings, conditional overexpression of FKBP12.6 abrogated triggered ventricular tachycardia (Gellen et al., 2008).

Other studies have failed to establish that PKA phosphorylation of RyR2-S2809 and dissociation of FKBP12.6 from RyR2 are associated with a leaky SR Ca^{2+} channel. Li, Kranias, Mignery, and Bers (2002) measured resting Ca^{2+} sparks in permeabilized ventricular myocytes isolated from WT mice and mice lacking SERCA-regulatory subunit, phospholamban. The finding that PKA-mediated phosphorylation increased Ca^{2+} spark amplitude and duration in WT but not phospholamban knockout myocytes suggested that the primary mechanism of PKA action causing alterations in Ca^{2+}-homeostasis during heart failure involves phospholamban phosphorylation, and thus SERCA activity. Jiang et al. (2002) investigated the SR Ca^{2+} handling properties of human failing hearts and in a tachycardia-induced canine model of heart failure. The amplitude and kinetics of the Ca^{2+} transients were reduced. However, RyR2s isolated from canine and human heart failure tissues displayed no major structural or functional differences compared with controls. Furthermore, phosphorylation of RyRs by PKA did not appear to dissociate FKBP12.6 from the RyR2s. Xiao, Tian, Jones, et al. (2007) showed that removal of FKBP12.6 neither altered the functional properties of RyR2 nor increased the susceptibility of FKBP12.6$^{-/-}$ mice to stress-induced ventricular arrhythmias. Moreover, deletion of the Ser2809 phosphorylation site did not alter the β-adrenergic response in whole hearts and isolated cardiomyocytes. Aortic-banded RyR2-S2809A and WT mice both developed cardiac hypertrophy in absence of PKA-mediated phosphorylation of RyR2 (Benkusky et al., 2007; MacDonnell et al., 2008).

In skeletal muscle, during exercise, hyperphosphorylation of RyR1-Ser2843 by PKA, *S*-nitrosylation (see below), loss of phosphodiesterase PDE4D3, and the RyR1 subunit FKBP12 have been linked to decreased muscle function (Bellinger et al., 2008). A small

molecule, S107 (whose action is discussed later in more detail), resulted in improved muscle function by stabilizing the RyR1–FKBP12 complex.

C. RyR Modulation by CaMKII-Mediated Phosphorylation

Phosphorylation of RyR2 by CaMKII has also been implicated in muscle diseases. Single channel recordings indicated that phosphorylation by CaMKII increases WT-RyR2 activity and Ca^{2+} sensitivity, but not of RyR2-S2815A that lacks the RyR2 CaMKII phosphorylation site (Wehrens, Lehnart, et al., 2004). Hain et al. (1995) proposed that phosphorylation of one subunit of the tetrameric RyR2 by endogenous CaMKII results in channel blockade by Mg^{2+}, whereas phosphorylation of all four subunits by exogenous CaMKII opens the channel. Transgenic mice that overexpressed the major cytosolic form of CaMKII ($CaMKII\delta_c$) had an increased phosphorylation of RyR2 and phospholamban, and died prematurely (Zhang et al., 2003). Ca^{2+} spark activities were enhanced in transgenic $CaMKII\delta_c$ overexpressing hearts despite reduced SR Ca^{2+} content (Maier et al., 2003). This implied that CaMKII-mediated RyR2 phosphorylation results in the formation of leaky SR Ca^{2+} channels. In support of this view, creation of a genetic mouse model of CaMKII kinase inhibition protected the heart against excessive β adrenergic stimulation and myocardial infarction (Zhang et al., 2005). An opposing view is that acute overexpression of constitutively active CaMKII phosphorylates RyR2 and decreases the rate of local Ca^{2+} release events (Ca^{2+} sparks) and Ca^{2+} waves in cultured rat cardiomyocytes (Yang et al., 2007). In more recent studies using transgenic and KO mouse models, increased CaMKII activity contributed to increased SR leak, cardiac arrhythmogenesis and heart failure, whereas CaMKII deficiency reduced isoproterenol-induced arrhythmias and progression to heart failure (Ling et al., 2009; Sag et al., 2009). Chelu et al. (2009) reported that rapid atrial pacing unmasked increased vulnerability to atrial fibrillation in a genetic RyR2 gain-of-function mouse model compared to WT. This was ascribed to increased SR Ca^{2+} leak due to enhanced CaMKII-mediated phosphorylation of RyR2.

D. Protective Effects of 1,4-Benzothiazepine Derivatives JTV519 (K201) and S107

JTV519 (also known as K201) is a 1,4-benzothiazepine derivative that has a strong cardioprotective effect. This results from presumably inhibiting a broad spectrum of ion channel function (Hasumi, Matsuda, Shimamoto, Hata, & Kaneko, 2007). Yano et al. (2003) found that JTV519 reduced SR Ca^{2+} leak and improved cardiac function in dogs subjected to 4 weeks of chronic right ventricular pacing by minimizing RyR2 hyperphosphorylation and stabilizing the RyR2/FKBP12.6 complex. Single channel recordings showed that JTV519 stabilizes the closed state of RyR2 by promoting FKBP12.6 binding (Wehrens et al., 2004). The ineffectiveness of JTV519 to prevent exercise-induced ventricular tachyarrhythmias and death in $FKBP12.6^{-/-}$ mice suggested that binding of FkBP12.6 to RyR2 is a critical step for the drug having a beneficial effect. In two recent studies, the RyR2-specific derivative of JTV519, S107, enhanced the binding of FKBP12.6 to CPVT-associated RyR2-R2474S mutant (Lehnart et al., 2008), and in a dystrophic mouse model binding of FKBP12 to the hypernitrosylated RyR1 (Bellinger et al., 2009). In both cases, inhibition of channel leak was observed. At variance, JTV519 suppressed spontaneous Ca^{2+} release in FK506 treated HEK 293 cells and inhibited [^{3}H]-ryanodine binding to RyR2-N4104K linked to ventricular tachycardia, in the absence of FKPB12.6 (Hunt et al., 2007). Blayney, Jones, Griffiths, and Lai (2009) recently reported that both PKA-mediated phosphorylation and JTV519 change the affinity of FKBP12.6 binding to RyR2. Both reduced binding affinity to the closed RyR2 at 10^{-8} M Ca^{2+}, whereas a moderate increase in binding affinity was observed to the partially open channel at 10^{-3} M Ca^{2+}. Matsuzaki and colleagues (Oda et al., 2005; Tateishi et al., 2009) provided an explanation for the observation that JTV519 and its derivative S107 restore normal function for both phosphorylated and nitrosylated channels. These authors reported that disrupted (unzipped)

domain interactions between the N-terminal and central domains of RyR2 are responsible for diastolic Ca^{2+} leak in failing hearts. Stabilization of these interactions by JTV519 improved cardiac function.

IV. RYR MODULATION BY REACTIVE OXYGEN AND NITROGEN SPECIES

A. RyRs and Reactive Oxygen Species

Active muscle produces reactive nitrogen and oxygen species that modulate contraction and relaxation (Ji, 2000; Reid, 1996; Stamler & Meissner, 2001). During strenuous exercise or short episodes of ischemia followed by reoxygenation, increased levels of reactive oxygen species impose oxidative stress. Reactive oxygen species can be formed by several mechanisms. These include the mitochondrial electron transport chain, xanthine oxidase, and NAD(P)H oxidases. Cells respond to excessive reactive oxygen species via the action of superoxide dismutase which converts superoxide ($O_2^{\bullet -}$) to O_2 and H_2O_2, and catalase and glutathione peroxidase which decompose H_2O_2.

Early studies showed that redox active reagents such as heavy metals, *N*-ethylmaleimide, diamide, $O_2^{\bullet -}$, and H_2O_2 modulate RyR activity (Abramson & Salama, 1989; Aghdasi, Zhang, Wu, Reid, & Hamilton, 1997; Anzai et al., 1998; Kawakami & Okabe, 1998; Oba, Ishikawa, & Yamaguchi, 1998). RyRs are good targets for redox active species because they contain a large number of thiols. The tetrameric mammalian skeletal and cardiac RyRs have 404 and 364 cysteines, with 100 and 89 cysteines per 560 kDa subunit, and 1 and 2 per FK506 binding protein, respectively.

The number of free thiols in purified RyR1 was determined using the lipophilic, thiol-specific probe, monobromobimane (Eu, Sun, Xu, Stamler, & Meissner, 2000). Nearly half of the 404 cysteines in the tetrameric RyR1 channel complex were labeled, that is, free in the presence of 5 mM reduced glutathione (GSH) at pO_2 ~ 10 mmHg. An increase in oxygen tension from ~10 mmHg (simulating tissue pO_2) to ambient air (pO_2 ~ 150 mmHg) in the presence of 5 mM GSH, resulted in loss of six to eight free thiols/RyR1 subunit without appreciably changing RyR1 activity. Substitution of GSH with oxidized glutathione (GSSG) at pO_2 ~ 10 mmHg or ambient air reduced the number of free thiols/RyR1 subunit and resulted in a large increase in RyR1 activity. The number of free thiols in RyR2-enriched fractions in the presence of 5mMGSH was 50 per mg protein at pO_2 ~ 150 mmHg and 58 per mg protein ~10 mmHg (Sun et al., 2008). In the presence of 5 mM GSSG, a lower number of free cysteines were measured. There existed a good correlation between RyR2 activity and free RyR2 thiol content; however, because RyR2 was only partially purified, the exact number of glutathione and pO_2-sensitive thiols in native RyR2 remains to be determined. The results suggest thatRyR1 and RyR2 have functional thiols that respond to the cells' pO_2 and GSH/GSSG redox state.

Additional cellular parameters influence the redox state of RyRs. Single channel measurements have shown that RyR1 responds to changes in cytosolic and SR luminal glutathione redox potential (Feng, Liu, Allen, & Pessah, 2000). Micromolar activating Ca^{2+} concentrations lowered the redox state of RyR1 and favored channel opening, whereas inhibitory concentrations of Ca^{2+} and Mg^{2+} had opposite effects (Xia, Stangler, & Abramson, 2000). The observation that channel activators such as caffeine may act as electron acceptors, whereas inhibitors such as tetracaine may act as electron donors, suggested that nonthiol channel modulators regulate channel activity by shifting the thiol–disulfide ratio in the RyRs (Marinov, Olojo, Xia, & Abramson, 2007). RyR1-associated proteins have been shown to also affect receptor redox state. Channel closing unmasked the presence of hyperreactive cysteine residues in both RyR1 and triadin (Liu, Abramson, Zable, & Pessah, 1994), and SepN1, a selenoprotein, maintained the receptor's normal sensitivity

to redox active species (Jurynec et al., 2008). Varied regulation of RyRs by plasmalemmal- and SR-associated NAD(P)H oxidases has been described (Espinosa et al., 2006; Hidalgo, Sanchez, Barrientos, & Aracena-Parks, 2006; Sanchez et al., 2008; Xia, Webb, Gnall, Cutler, & Abramson, 2003; Zima & Blatter, 2006).

A number of redox reactive cysteines among the 100 cysteines per RyR1 subunit have been identified, although the functional significance of specific cysteines remains elusive. Voss, Lango, Ernst-Russell, Morin, and Pessah (2004) determined by mass spectrometric analysis seven hyperreactive RyR1 cysteines (Cys-1040, 1303, 2436, 2565, 2606, 2611, and 3635) that were selectively labeled by 7-diethylamino-3-(4′-maleimidylphenyl)-4-methylcoumarin(CPM) under conditions that favored channel closing. Petrotchenko et al. (2006) identified by mass spectrometry one cysteine in RyR1 (Cys2327) that responded to a change in glutathione redox state. Aracena-Parks et al. (2006) showed that nine RyR1 cysteines (Cys-36, 315, 811, 906, 1591, 2326, 2363, 3193, and 3635) are endogenously modified and another three (Cys-253, 1040, and 1303) are modified by exogenous reactive oxygen and nitrogen molecules.

B. Regulation of RyRs by Nitric Oxide and Related Molecules

Nitric oxide (NO) is a ubiquitous regulator of cellular function and physiological modulator of cardiac and skeletal muscle excitation–contraction coupling. *S*-Nitrosylation of cardiac L-type Ca^{2+} channel (Sun et al., 2006) and RyRs (see below) have been described. Mammalian tissues express four isoforms of nitric oxide synthase: endothelial (eNOS), neuronal (nNOS), inducible (iNOS), and mitochondrial (mNOS) forms. In normal skeletal and cardiac muscle, the predominant isoforms, nNOS and eNOS, are targeted to the sarcolemma via interaction with the dystrophin-associated glycoprotein complex (Brenman, Chao, Xia, Aldape, & Bredt, 1995) and the caveolae structural protein caveolin-3 (Feron et al., 1996), respectively. Immunoelectron microscopy showed nNOS labeling in isolated cardiac SR vesicles, whereas no labeling was detected in skeletal muscle SR vesicles (Xu, Huso, Dawson, Bredt, & Becker, 1999). This suggested that NO regulates cardiac function by spatial confinement of nNOS and eNOS isoforms, *S*-nitrosylating RyR2 and plasmalemmal L-type Ca^{2+} channel, respectively. Studies with $eNOS^{-/-}$ and $nNOS^{-/-}$ mice support the idea that nNOS has a specific role in regulating SR Ca^{2+} release (Barouch et al., 2002). Although both $NOS^{-/-}$ mice developed age-related hypertrophy, only $eNOS^{-/-}$ mice were hypertensive. $nNOS^{-/-}$ mice exhibited a reduced ionotropic response, whereas $eNOS^{-/-}$ mice had enhanced contractility due to increased SR Ca^{2+} release. iNOS is absent or very low in normal heart but may increase in concentration, depending on the disease state (Arstall, Sawyer, Fukazawa, & Kelly, 1999).

NO exerts its cellular effects via cGMP-dependent and -independent pathways (Stamler & Hausladen, 1998). In the cGMP-dependent pathway, binding of NO to the heme group of guanylate cyclase increases the production of intracellular cGMP and activation of cGMP-dependent kinase, which was reported to phosphorylate RyR2 (Takasago et al., 1991). NO operates independently of cGMP through *S*-nitrosylation of proteins, most often at a single cysteine in acid–base or hydrophobic motifs (Hess, Matsumoto, Nudelman, & Stamler, 2001). Modification of RyR may involve other species, such as peroxynitrite ($OONO^-$), which is formed via a reaction of NO with superoxide. Peroxynitrite extensively oxidizes the RyRs (Sun, Xu, Eu, Stamler, & Meissner, 2001; Xu, Eu, Meissner, & Stamler, 1998) and has been implicated in myocardial injury and loss of RyR2 activity in the postischemic heart (Tang et al., 2009; Wang & Zweier, 1996).

Both RyR1 (Eu et al., 2000) and RyR2 (Xu et al., 1998) are endogenously *S*-nitrosylated, suggesting that NO is a physiological modulator of skeletal and cardiac muscle excitation–contraction coupling. In *in vitro* studies, NO or NO-related species activated or inhibited

RyRs depending on donor concentration, membrane potential, the presence of channel agonists, and other sulfhydryl modifying reagents (Aghdasi, Reid, & Hamilton, 1997; Hart & Dulhunty, 2000; Meszaros, Minarovic, & Zahradnikova, 1996; Suko, Drobny, & Hellmann, 1999; Zahradnikova, Minarovic, Venema, & Meszaros, 1997). Low physiological concentrations of NO *S*-nitrosylated and activated RyR1 at tissue pO_2 (~10 mmHg) but not in ambient air (pO_2 ~ 150 mmHg) (Eu et al., 2000). Changes in oxygen tension oxidized/reduced as many as six to eight thiols in each RyR1 subunit, which may explain the responsiveness of RyR1 to NO at tissue pO_2 but not ambient air. In intact muscle, NO modulated the O_2 tension dependence of SR Ca^{2+} release and contractility (Eu et al., 2003). The results suggest that RyR1 is an O_2 and NO sensing molecule, an idea questioned by Cheong, Tumbev, Stoyanovsky, and Salama (2005). These investigators found that NO did not activate RyR1 under a range of pO_2. Like RyR1, RyR2 activity is dependent on pO_2 (Sun et al., 2008). However, unlike RyR1, RyR2 was not effectively activated and *S*-nitrosylated by NO. RyR2 was nonetheless modified and activated by *S*-nitrosoglutathione (GSNO) and $ONOO^-$.

3-Morpholinosydnonimine (SIN-1) (which generates peroxynitrite, $ONOO^-$), NOC12 (which generates a variety of reactive nitrogen oxides), and GSNO activate RyR1 independently of oxygen tension. NOC-12 activated by *S*-nitrosylation (Sun, Xu, Eu, Stamler, &Meissner, 2003), SIN-1 by oxidation of thiols (Sun, Xu, et al., 2001b), and GSNO by *S*-nitrosylation/oxidation (Sun et al., 2003) and *S*-glutathionylation (Aracena, Sanchez, Donoso, Hamilton, & Hidalgo, 2003). Peroxynitrite also modifies protein tyrosines (Souza, Peluffo, & Radi, 2008). The isolation of 3-nitrotyrosine containing fragments from type 2a Ca^{2+} ATPase (SERCA2a) and type 3 RyR (RyR3) isoforms in aging skeletal muscle was reported (Kanski, Hong, & Schoneich, 2005).

Site-directed mutagenesis studies demonstrated that at physiological O_2 concentrations, NO specifically *S*-nitrosylated Cys3635 out of ~50 free cysteines/RyR1 subunit (Sun, Xin, Eu, Stamler, & Meissner, 2001). Cys3635 is located in the CaM binding domain of RyR1 (Porter Moore, Zhang, & Hamilton, 1999; Yamaguchi, Xin, & Meissner, 2001), which provides an explanation that NO transduces its functional effect only in the presence of calmodulin. In contrast, activation of RyR1 by GSNO was independent of C3635 and calmodulin. Likewise, the corresponding RyR2 cysteine (C3602) was not required for RyR2 activation by GSNO (Sun et al., 2008).

C. RyR Oxidation and *S*-Nitrosylation in Normal and Diseased Muscle

Given the large number of free thiols, it is not surprising that abnormal modulation of RyRs by redox active molecules is implicated in muscle diseases. Durham et al. (2008) studied the role of RyR1-Y522S mutation in a knockin mouse model. In humans, the mutation is associated with malignant hyperthermia and a high incidence of central cores (Quane et al., 1994). Heterozygous expression of RyR1-Y522S increased SR Ca^{2+} leak, which led to the increased production of reactive nitrogen species. Increased *S*-nitrosylation of RyR1 further enhanced SR Ca^{2+} leak and resulted in increased susceptibility to heat-induced death. In fast-twitch fibers, the skeletal muscle-specific neuronal nitric oxide synthase (nNOSμ) is localized to the sarcolemma via interaction with the dystrophin-associated glycoprotein complex (Brenman et al., 1995). In mdx mice and in humans with Duchenne muscular dystrophy (the most common form of muscular dystrophy) disruption of the dystrophin-associated glycoprotein complex decreased the expression of nNOSμ. Despite a decreased nNOSμ expression, RyR1 was hypernitrosylated in mdx mice (Bellinger et al., 2009). This was due to increased expression and formation of a complex of inducible nitric oxide synthase (iNOS) with RyR1. Increased *S*-nitrosylation of RyR1 correlated with dissociation of FKBP12 from RyR1 and formation of leaky Ca^{2+} channels. S107, a compound previously shown by the authors to enhance the binding affinity of FKBP to hyperphosphorylated RyRs

(Bellinger et al., 2008), enhanced muscle performance by increasing FKBP binding affinity to RyR1. SR Ca^{2+} leak and muscle damage were reduced. Interestingly, no increases in PKA-mediated phosphorylation of RyR1 at Ser2843 were observed in mdx mice. Thus, S107 may stabilize the FKBP–RyR1 complex under conditions where RyR1 is modified by very different mechanisms.

Aberrant oxidation and *S*-nitrosylation of RyR2 have been implicated in ischemic and failing hearts. Physical association of xanthine oxidoreductase (Khan et al., 2004) and nNOS (Khan et al., 2004; Xu et al., 1999) suggests that RyR2 is subjected to modifications by NO and superoxide ($O_2^{\bullet-}$) and their derivatives. NO can form HNO, an 1 electron reduction product of NO, reacts with $O_2^{\bullet-}$ to form peroxynitrite ($OONO^-$), and reacts with glutathione to generate GSNO. All three products activate RyR2 (Cheong, Tumbev, Abramson, Salama, & Stoyanovsky, 2005; Tocchetti et al., 2007; Xu et al., 1998). Adding to the complexity of the potential regulation ofRyR2bymultiple oxygen and nitrogen reactive species, a translocation of nNOS from the SR to the sarcolemmal caveolae was observed in a rat model of heart failure (Bendall et al., 2004) and failing human hearts (Damy et al., 2004). Displacement of nNOS from RyR2 removes inhibition of xanthine oxidoreductase by NO, which can increase $O_2^{\bullet-}$ production and oxidative stress in failing hearts.

In the ischemic heart, increased NO production was associated with reduced myocardial contractility (Node et al., 1996; Zweier, Wang, & Kuppusamy, 1995). In the postischemic heart during the early period of reflow, NO production increased and reacted with superoxide to form peroxynitrite ($OONO^-$), which caused amino acid nitration and cellular injury (Wang & Zweier, 1996). Other studies have suggested that NO has a cardioprotective role. Physiologically relevant concentrations of peroxynitrite protected against myocardial reperfusion injury (Lefer et al., 1997). Cardiomyocyte-specific overexpression of eNOS limited left ventricular dysfunction after myocardial infarction (Janssens et al., 2004). Preconditioning and application of GSNO resulted in a similar pattern in protein *S*-nitrosylation and cardiac protection against ischemia/reperfusion (Sun, Morgan, Shen, Steenbergen, & Murphy, 2007). This suggested that *S*-nitrosylation of protein thiols protects cells from further oxidative damage. Although it is likely that RyR2 is modified in the ischemic/reperfused heart, the redox modifications that may occur are unclear.

Gonzalez, Beigi, Treuer, and Hare (2007) reported that diastolic Ca^{2+} levels increased in $nNOS^{-/-}$ but not $eNOS^{-/-}$ mice. nNOS elimination was associated with decreased *S*-nitrosylation, increased oxidation of RyR2 and SR Ca^{2+} leak, and arrhythmogenesis in cardiomyocytes. FKBP12.6 binding and phosphorylation of RyR2 were not altered in $nNOS^{-/-}$ mice. On the other hand, in hearts of mdx mice an increased *S*-nitrosylation and partial dissociation of FKBP12.6 from RyR2 was associated with a diastolic Ca^{2+} leak and arrhythmias (Fauconnier et al., 2010). S107 stabilized the RyR2/FKBP16.6 complex, reduced the SR Ca^{2+} leak, and prevented arrhythmias *in vivo*, without affecting the *S*-nitrosylation state of RyR2. The possibility of increased oxidation of RyR2 in mdx mice was not described by Fauconnier et al. (2010) but may have contributed to the formation of the SR Ca^{2+} leak, because treatment of cardiac and skeletal muscle SR membranes with the sulfhydryl oxidizing agents, H_2O_2 and diamide, diminished FKBP12.6 binding (Zissimopoulos, Docrat, & Lai, 2007). Consistent with this finding, Terentyev et al. (2008) observed that oxidizing agents increased SR Ca^{2+} leak in cardiomyocytes from normal hearts. In cardiomyocytes isolated from a chronic model of heart failure, reduced/oxidized glutathione ratio was decreased. The reduced level of free RyR2 thiols and enhanced SR Ca^{2+} leak were partially restored to normal levels by treating heart failure cardiomyocytes with sulfhydryl reducing agents.

V. CONCLUSIONS

Although recent work improved our understanding of the interaction of protein kinases and redox active molecules with the RyRs, their mechanisms in modulating Ca^{2+} handling in normal and dysfunctional muscle remain controversial. The third mammalian RyR isoform, RyR3, contains putative phosphorylation and redox active sites; however, coexpression with the other mammalian RyR isoforms has hindered establishment of a specific cellular role. Importantly, progress has been made in the development of drugs that stabilize RyR activity in dysfunctional muscle. However, their mechanism of action remains elusive. One major limitation in studying the RyRs is that their solution structure has not been solved. Cellular studies of protein kinases and redox active molecules are complicated by multiple interactions of the RyRs and potential changes in additional signaling mechanisms and transport proteins.

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References

Abramson JJ, Salama G. Critical sulfhydryls regulate calcium release from sarcoplasmic reticulum. Journal of Bioenergetics and Biomembranes. 1989; 21:283–294. [PubMed: 2666411]

Aghdasi B, Reid MB, Hamilton SL. Nitric oxide protects the skeletal muscle Ca^{2+} release channel from oxidation induced activation. The Journal of Biological Chemistry. 1997; 272:25462–25467. [PubMed: 9325258]

Aghdasi B, Zhang JZ, Wu Y, Reid MB, Hamilton SL. Multiple classes of sulfhydryls modulate the skeletal muscle Ca^{2+} release channel. The Journal of Biological Chemistry. 1997; 272:3739–3748. [PubMed: 9013631]

Anzai K, Ogawa K, Kuniyasu A, Ozawa T, Yamamoto H, Nakayama H. Effects of hydroxyl radical and sulfhydryl reagents on the open probability of the purified cardiac ryanodine receptor channel incorporated into planar lipid bilayers. Biochemical and Biophysical Research Communications. 1998; 249:938–942. [PubMed: 9731240]

Aracena P, Sanchez G, Donoso P, Hamilton SL, Hidalgo C. S-glutathionylation decreases Mg^{2+} inhibition and S-nitrosylation enhances Ca^{2+} activation of RyR1 channels. The Journal of Biological Chemistry. 2003; 278:42927–42935. [PubMed: 12920114]

Aracena-Parks P, Goonasekera SA, Gilman CP, Dirksen RT, Hidalgo C, Hamilton SL. Identification of cysteines involved in S-nitrosylation, S-glutathio-nylation, and oxidation to disulfides in ryanodine receptor type 1. The Journal of Biological Chemistry. 2006; 281:40354–40368. [PubMed: 17071618]

Arstall MA, Sawyer DB, Fukazawa R, Kelly RA. Cytokine-mediated apoptosis in cardiac myocytes. The role of inducible nitric oxide synthase induction and peroxynitrite generation. Circulation Research. 1999; 85:829–840. [PubMed: 10532951]

Barouch LA, Harrison RW, Skaf MW, Rosas GO, Cappola TP, Kobeissi ZA, et al. Nitric oxide regulates the heart by spatial confinement of nitric oxide synthase isoforms. Nature. 2002; 416:337–339. [PubMed: 11907582]

Bellinger AM, Reiken S, Carlson C, Mongillo M, Liu X, Rothman L, et al. Hypernitrosylated ryanodine receptor calcium release channels are leaky in dystrophic muscle. Natural Medicines. 2009; 15:325–330.

Bellinger AM, Reiken S, Dura M, Murphy PW, Deng SX, Landry DW, et al. Remodeling of ryanodine receptor complex causes “leaky” channels, a molecular mechanism for decreased exercise capacity. Proceedings of the National Academy of Sciences of the United States of America. 2008; 105:2198–2202. [PubMed: 18268335]

Bendall JK, Damy T, Ratajczak P, Loyer X, Monceau V, Marty I, et al. Role of myocardial neuronal nitric oxide synthase-derived nitric oxide in beta-adrenergic hyporesponsiveness after myocardial infarction-induced heart failure in rat. Circulation. 2004; 110:2368–2375. [PubMed: 15466641]

Benkusky NA, Weber CS, Scherman JA, Farrell EF, Hacker TA, John MC, et al. Intact beta-adrenergic response and unmodified progression toward heart failure in mice with genetic ablation of a major protein kinase A phosphorylation site in the cardiac ryanodine receptor. Circulation Research. 2007; 101:819–829. [PubMed: 17717301]

Bers DM. Cardiac excitation-contraction coupling. Nature. 2002; 415:198–205. [PubMed: 11805843]

Blayney LM, Jones JL, Griffiths J, Lai FA. A mechanism of ryanodine receptor modulation by FKBP12/12.6, protein kinase A, and K201. Cardiovascular Research. 2009; 85:68–78. [PubMed: 19661110]

Brenman JE, Chao DS, Xia H, Aldape K, Bredt DS. Nitric oxide synthase complexed with dystrophin and absent from skeletal muscle sarcolemma in Duchenne muscular dystrophy. Cell. 1995; 82:743–752. [PubMed: 7545544]

Brillantes AB, Ondrias K, Scott A, Kobrinsky E, Ondriasova E, Moschella MC, et al. Stabilization of calcium release channel (ryanodine receptor) function by FK506-binding protein. Cell. 1994; 77:513–523. [PubMed: 7514503]

Callaway C, Seryshev A, Wang JP, Slavik KJ, Needleman DH, Cantu CI, et al. Localization of the high and low affinity [^{3}H]ryanodine binding sites on the skeletal muscle Ca^{2+} release channel. The Journal of Biological Chemistry. 1994; 269:15876–15884. [PubMed: 8195243]

Carter S, Coyler J, Sitsapesan R. Maximum phosphorylatioin of the cardiac ryanodine receptor at serine-2809 by protein kinase A produces unique modifications to channel gating and conductance not observed at lower levels to phosphorylation. Circulation Research. 2006; 98:1506–1513. [PubMed: 16709901]

Chelu MG, Sarma S, Sood S, Wang S, van Oort RJ, Skapura DG, et al. Calmodulin kinase II-mediated sarcoplasmic reticulum Ca^{2+} leak promotes atrial fibrillation in mice. The Journal of Clinical Investigation. 2009; 119:1940–1951. [PubMed: 19603549]

Cheong E, Tumbev V, Abramson J, Salama G, Stoyanovsky DA. Nitroxyl triggers Ca^{2+} release from skeletal and cardiac sarcoplasmic reticulum by oxidizing ryanodine receptors. Cell Calcium. 2005; 37:87–96. [PubMed: 15541467]

Cheong E, Tumbev V, Stoyanovsky D, Salama G. Effects of pO(2) on the activation of skeletal muscle ryanodine receptors by NO, A cautionary note. Cell Calcium. 2005; 38:481–488. [PubMed: 16099502]

Currie S, Loughrey CM, Craig MA, Smith GL. Calcium/calmodulin-dependent protein kinase IIdelta associates with the ryanodine receptor complex and regulates channel function in rabbit heart. The Biochemical Journal. 2004; 277:357–366. [PubMed: 14556649]

Damy T, Ratajczak P, Shah AM, Camors E, Marty I, Hasenfuss G, et al. Increased neuronal nitric oxide synthase-derived NO production in the failing human heart. Lancet. 2004; 363:1365–1367. [PubMed: 15110495]

Du GG, Avila G, Sharma P, Khanna VK, Dirksen RT, MacLennan DH. Role of the sequence surrounding predicted transmembrane helix M4 in membrane association and function of the Ca^{2+} release channel of skeletal muscle sarcoplasmic reticulum (ryanodine receptor isoform 1). The Journal of Biological Chemistry. 2004; 279:37566–37574. [PubMed: 15226293]

Du GG, Sandhu B, Khanna VK, Guo XH, MacLennan DH. Topology of the Ca^{2+} release channel of skeletal muscle sarcoplasmic reticulum (RyR1). Proceedings of the National Academy of Sciences of the United States of America. 2002; 99:16725–16730. [PubMed: 12486242]

Dulhunty AF, Laver D, Curtis SM, Pace S, Haarmann C, Gallant EM. Characteristics of irreversible ATP activation suggest that native skeletal ryanodine receptors can be phosphorylated via an endogenous CaMKII. Biophysical Journal. 2001; 81:3240–3252. [PubMed: 11720989]

Durham WJ, Aracena-Parks P, Long C, Rossi AE, Goonasekera SA, Boncompagni S, et al. RyR1 S-nitrosylation underlies environmental heat stroke and sudden death in Y522S RyR1 knockin mice. Cell. 2008; 133:53–65. [PubMed: 18394989]

Espinosa A, Leiva A, Pena M, Muller M, Debandi A, Hidalgo C, et al. Myotube depolarization generates reactive oxygen species through NAD(P)H oxidase; ROS-elicited Ca^{2+} stimulates ERK, CREB, early genes. Journal of Cellular Physiology. 2006; 209:379–388. [PubMed: 16897752]

Eu JP, Hare JM, Hess DT, Skaf M, Sun J, Cardenas-Navina I, et al. Concerted regulation of skeletal muscle contractility by oxygen tension and endogenous nitric oxide. Proceedings of the National Academy of Sciences of the United States of America. 2003; 100:15229–15234. [PubMed: 14645704]

Eu JP, Sun J, Xu L, Stamler JS, Meissner G. The skeletal muscle calcium release channel, coupled O_2 sensor and NO signaling functions. Cell. 2000; 102:499–509. [PubMed: 10966111]

Fabiato A. Calcium-induced release of calcium from the cardiac sarcoplasmic reticulum. The American Journal of Physiology. 1983; 245:C1–C14. [PubMed: 6346892]

Fauconnier J, Thireau J, Reiken S, Cassan C, Richard S, Matecki S, et al. Leaky RyR2 trigger ventricular arrhythmias in Duchenne muscular dystrophy. Proceedings of the National Academy of Sciences of the United States of America. 2010; 107:1559–1564. [PubMed: 20080623]

Feng W, Liu G, Allen PD, Pessah IN. Transmembrane redox sensor of ryanodine receptor complex. The Journal of Biological Chemistry. 2000; 275:35902–35907. [PubMed: 10998414]

Feron O, Belhassen L, Kobzik L, Smith TW, Kelly RA, Michel T. Endothelial nitric oxide synthase targeting to caveolae. Specific interactions with caveolin isoforms in cardiac myocytes and endothelial cells. The Journal of Biological Chemistry. 1996; 271:22810–22814. [PubMed: 8798458]

Fill M, Copello JA. Ryanodine receptor calcium release channels. Physiological Reviews. 2002; 82:893–922. [PubMed: 12270947]

Franzini-Armstrong C, Protasi F. Ryanodine receptors of striated muscles, a complex channel capable of multiple interactions. Physiological Reviews. 1997; 77:699–729. [PubMed: 9234963]

Furuichi T, Furutama D, Hakamata Y, Nakai J, Takeshima H, Mikoshiba K. Multiple types of ryanodine receptor/Ca^{2+} release channels are differentially expressed in rabbit brain. The Journal of Neuroscience. 1994; 14:4794–4805. [PubMed: 8046450]

Gellen B, Fernandez-Velasco M, Briec F, Vinet L, LeQuang K, Rouet-Benzineb P, et al. Conditional FKBP12.6 overexpression in mouse cardiac myocytes prevents triggered ventricular tachycardia through specific alterations in excitation-contraction coupling. Circulation. 2008; 117:1778–1786. [PubMed: 18378612]

Gonzalez DR, Beigi F, Treuer AV, Hare JM. Deficient ryanodine receptor S-nitrosylation increases sarcoplasmic reticulum calcium leak and arrhythmogenesis in cardiomyocytes. Proceedings of the National Academy of Sciences of the United States of America. 2007; 104:20612–20617. [PubMed: 18077344]

Hain J, Onoue H, Mayrleitner M, Fleischer S, Schindler H. Phosphorylation modulates the function of the calcium release channel of sarcoplasmic reticulum from cardiac muscle. The Journal of Biological Chemistry. 1995; 270:2074–2081. [PubMed: 7836435]

Hart JD, Dulhunty AF. Nitric oxide activates or inhibits skeletal muscle ryanodine receptors depending on its concentration, membrane potential and ligand binding. The Journal of Membrane Biology. 2000; 173:227–336. [PubMed: 10667918]

Hasumi H, Matsuda R, Shimamoto K, Hata Y, Kaneko N. K201, a multi-channel blocker, inhibits clofilium-induced torsades de pointes and attenuates an increase in repolarization. European Journal of Pharmacology. 2007; 555:54–60. [PubMed: 17112502]

Hess DT, Matsumoto A, Nudelman R, Stamler JS. S-nitrosylation, spectrum and specificity. Nature Cell Biology. 2001; 3:E46–E49.

Hidalgo C, Sanchez G, Barrientos G, Aracena-Parks P. A transverse tubule NOX activity stimulates calcium release from isolated triads via RYR1 S-Glutathionylation. The Journal of Biological Chemistry. 2006; 281:26473–26482. [PubMed: 16762927]

Hohenegger M, Suko J. Phosphorylation of the purified cardiac ryanodine receptor by exogenous and endogenous protein kinases. The Biochemical Journal. 1993; 296:303–308. [PubMed: 8257417]

Hunt DJ, Jones PP, Wang R, Chen W, Bolstad J, Chen K, et al. K201 (JTV519) suppresses spontaneous Ca^{2+} release and [^{3}H]ryanodine binding to RyR2 irrespective of FKBP12.6 association. The Biochemical Journal. 2007; 404:431–438. [PubMed: 17313373]

Janssens S, Pokreisz P, Schoonjans L, Pellens M, Vermeersch P, Tjwa M, et al. Cardiomyocyte-specific overexpression of nitric oxide synthase 3 improves left ventricular performance and reduces compensatory hypertrophy after myocardial infarction. Circulation Research. 2004; 94:1256–1262. [PubMed: 15044322]

Ji, LL. Exercise-induced oxidative stress in the heart. In: Sen, CK.; Packer, L.; Hanninen, O., editors. Handbook of exercise and oxygen toxity. Amsterdam, The Netherlands: Elsevier Science; 2000. p. 689-712.

Jiang MT, Lokuta AJ, Farrell EF, Wolff MR, Haworth RA, Valdivia HH. Abnormal Ca^{2+} release, but normal ryanodine receptors, in canine and human heart failure. Circulation Research. 2002; 91:1015–1022. [comment]. [PubMed: 12456487]

Jones PP, Meng X, Xiao B, Cai S, Bolstad J, Wagenknecht T, et al. Localization of PKA phosphorylation site, Ser(2030), in the three-dimensional structure of cardiac ryanodine receptor. The Biochemical Journal. 2008; 410:261–270. [PubMed: 17967164]

Jurynec MJ, Xia R, Mackrill JJ, Gunther D, Crawford T, Flanigan KM, et al. Selenoprotein N is required for ryanodine receptor calcium release channel activity in human and zebrafish muscle. Proceedings of the National Academy of Sciences of the United States of America. 2008; 105:12485–12490. [PubMed: 18713863]

Kanski J, Hong SJ, Schoneich C. Proteomic analysis of protein nitration in aging skeletal muscle and identification of nitrotyrosine-containing sequences in vivo by nanoelectrospray ionization tandem mass spectrometry. The Journal of Biological Chemistry. 2005; 280:24261–24266. [PubMed: 15851474]

Kawakami M, Okabe E. Superoxide anion radical-triggered Ca^{2+} release from cardiac sarcoplasmic reticulum through ryanodine receptor Ca^{2+} channel. Molecular Pharmacology. 1998; 53:497–503. [PubMed: 9495817]

Khan SA, Lee K, Minhas KM, Gonzalez DR, Raju SV, Tejani AD, et al. Neuronal nitric oxide synthase negatively regulates xanthine oxidoreductase inhibition of cardiac excitation-contraction coupling. Proceedings of the National Academy of Sciences of the United States of America. 2004; 101:15944–15948. [PubMed: 15486091]

Koch WJ. Genetic and phenotypic targeting of beta-adrenergic signaling in heart failure. Molecular and Cellular Biochemistry. 2004; 263:5–9. [PubMed: 15524162]

Lam E, Martin MM, Timerman AP, Sabers C, Fleischer S, Lukas T, et al. A novel FK506 binding protein can mediate the immunosuppressive effects of FK506 and is associated with the cardiac ryanodine receptor. The Journal of Biological Chemistry. 1995; 270:26511–26522. [PubMed: 7592869]

Lefer DJ, Scalia R, Campbell B, Nossuli T, Hayward R, Salamon M, et al. Peroxynitrite inhibits leukocyte-endothelial cell interactions and protects against ischemia-reperfusion injury in rats. The Journal of Clinical Investigation. 1997; 99:684–691. [PubMed: 9045871]

Lehnart SE, Mongillo M, Bellinger A, Lindegger N, Chen BX, Hsueh W, et al. Leaky Ca^{2+} release channel/ryanodine receptor 2 causes seizures and sudden cardiac death in mice. The Journal of Clinical Investigation. 2008; 118:2230–2245. [PubMed: 18483626]

Lehnart SE, Wehrens XH, Reiken S, Warrier S, Belevych AE, Harvey RD, et al. Phosphodiesterase 4D deficiency in the ryanodine-receptor complex promotes heart failure and arrhythmias. Cell. 2005; 123:25–35. [PubMed: 16213210]

Li Y, Kranias EG, Mignery GA, Bers DM. Protein kinase A phosphorylation of the ryanodine receptor does not affect calcium sparks in mouse ventricular myocytes. Circulation Research. 2002; 90:309–316. [PubMed: 11861420]

Ling H, Zhang T, Pereira L, Means CK, Cheng H, Gu Y, et al. Requirement for Ca^{2+}/calmodulin-dependent kinase II in the transition from pressure overload-induced cardiac hypertrophy to heart failure in mice. The Journal of Clinical Investigation. 2009; 119:1230–1240. [PubMed: 19381018]

Liu G, Abramson JJ, Zable AC, Pessah IN. Direct evidence for the existence and functional role of hyperreactive sulfhydryls on the ryanodine receptor-triadin complex selectively labeled by the coumarin maleimide 7-diethylamino-3-(4-maleimidylphenyl)-4-methylcoumarin. Molecular Pharmaceutics. 1994; 45:189–200.

MacDonnell SM, Garcia-Rivas G, Scherman JA, Kubo H, Chen X, Valdivia H, et al. Adrenergic regulation of cardiac contractility does not involve phoshorylation of cardiac ryanodine receptor at serine 2808. Circulation Research. 2008; 102:e65–e72. [PubMed: 18388322]

Maier LS, Zhang T, Chen L, DeSantiago J, Brown JH, Bers DM. Transgenic CaMKIIdeltaC overexpression uniquely alters cardiac myocyte Ca^{2+} handling, reduced SR Ca^{2+} load and activated SR Ca^{2+} release. Circulation Research. 2003; 92:904–911. [PubMed: 12676813]

Marinov BS, Olojo RO, Xia R, Abramson JJ. Non-thiol reagents regulate ryanodine receptor function by redox interactions that modify reactive thiols. Antioxidants Redox Signaling. 2007; 9:609–621. [PubMed: 17465884]

Marx SO, Gaburjakova J, Gaburjakova M, Henrikson C, Ondrias K, Marks AR. Coupled gating between cardiac calcium release channels (ryanodine receptors). Circulation Research. 2001; 88:1151–1158. [PubMed: 11397781]

Marx SO, Reiken S, Hisamatsu Y, Gaburjakova M, Gaburjakova J, Yang YM, et al. Phosphorylation-dependent regulation of ryanodine receptors, a novel role for leucine/isoleucine zippers. The Journal of Cell Biology. 2001; 153:699–708. [PubMed: 11352932]

Marx SO, Reiken S, Hisamatsu Y, Jayaraman T, Burkhoff D, Rosemblit N, et al. PKA phosphorylation dissociates FKBP12.6 from the calcium release channel (ryanodine receptor), defective regulation in failing hearts. Cell. 2000; 101:365–376. [PubMed: 10830164]

Meissner G. Ryanodine receptor/Ca^{2+} release channels and their regulation by endogenous effectors. Annual Review of Physiology. 1994; 56:485–508.

Meissner G. Regulation of mammalian ryanodine receptors. Frontiers in Bioscience. 2002; 7:d2072–d2080. [PubMed: 12438018]

Meng X, Xiao B, Cai S, Huang X, Li F, Bolstad J, et al. Three-dimensional localization of serine-2808, a phosphorylation site in cardiac ryanodine receptor. The Journal of Biological Chemistry. 2007; 282:25929–25939. [PubMed: 17606610]

Meszaros LG, Minarovic I, Zahradnikova A. Inhibition of the skeletal muscle ryanodine receptor calcium release channel by nitric oxide. FEBS Letters. 1996; 380:49–52. [PubMed: 8603745]

Michikawa T, Hamanaka H, Otsu H, Yamamoto A, Miyawaki A, Furuichi T, et al. Transmembrane topology and sites of N-glycosylation of inositol 1,4,5-trisphosphate receptor. The Journal of Biological Chemistry. 1994; 269:9184–9189. [PubMed: 8132655]

Neylon CB, Richards SM, Larsen MA, Agrotis A, Bobik A. Multiple types of ryanodine receptor/Ca^{2+} release channels are expressed in vascular smooth muscle. Biochemical and Biophysical Research Communications. 1995; 215:814–821. [PubMed: 7488046]

Node K, Kitakaze M, Kosaka H, Komamura K, Minamino T, Inoue M, et al. Increased release of NO during ischemia reduces myocardial contractility and improves metabolic dysfunction. Circulation. 1996; 93:356–364. [PubMed: 8548910]

Oba T, Ishikawa T, Yamaguchi M. Sulfhydryls associated with H_2O_2-induced channel activation are on luminal side of ryanodine receptors. The American Journal of Physiology. 1998; 274:C914–C921. [PubMed: 9575787]

Oda T, Yano M, Yamamoto T, Tokuhisa T, Okuda S, Doi M, et al. Defective regulation of interdomain interactions within the ryanodine receptor plays a key role in the pathogenesis of heart failure. Circulation. 2005; 111:3400–3410. [PubMed: 15967847]

Petrotchenko EV, Pasek D, Elms P, Dokholyan NV, Meissner G, Borchers CH. Combining fluorescence detection and mass spectrometric analysis for comprehensive and quantitative analysis of redox-sensitive cysteines in native membrane proteins. Analytical Chemistry. 2006; 78:7959–7966. [PubMed: 17134128]

Porter Moore C, Zhang JZ, Hamilton SL. A role for cysteine 3635 ofRYR1 in redox modulation and calmodulin binding. The Journal of Biological Chemistry. 1999; 274:36831–36834. [PubMed: 10601232]

Quane KA, Keating KE, Healy JM, Manning BM, Krivosic-Horber R, Krivosic I, et al. Mutation screening of the RYR1 gene in malignant hyperthermia, detection of a novel Tyr to Ser mutation in a pedigree with associated central cores. Genomics. 1994; 23:236–239. [PubMed: 7829078]

Reid MB. Reactive oxygen and nitric oxide in skeletal muscle. News in Physiological Sciences. 1996; 11:114–121.

Reiken S, Gaburjakova M, Guatimosim S, Gomez AM, D'Armiento J, Burkhoff D, et al. Protein kinase A phosphorylation of the cardiac calcium release channel (ryanodine receptor) in normal and failing hearts. Role of phosphatases and response to isoproterenol. The Journal of Biological Chemistry. 2003; 278:444–453. [PubMed: 12401811]

Reiken S, Lacampagne A, Zhou H, Kherani A, Lehnart SE, Ward C, et al. PKA phosphorylation activates the calcium release channel (ryanodine receptor) in skeletal muscle, defective regulation in heart failure. The Journal of Cell Biology. 2003; 160:919–928. [PubMed: 12629052]

Rios E, Pizarro G. Voltage sensor of excitation-contraction coupling in skeletal muscle. Physiological Reviews. 1991; 71:849–908. [PubMed: 2057528]

Rodriguez P, Bhogal MS, Colyer J. Stoichiometric phosphorylation of cardiac ryanodine receptor on serine 2809 by calmodulin-dependent kinase II and protein kinase A. The Journal of Biological Chemistry. 2003; 278:38593–38600. [PubMed: 14514795]

Ruehr ML, Russell MA, Ferguson DG, Bhat M, Ma J, Damron DS, et al. Targeting of protein kinase A by muscle A kinase-anchoring protein (mAKAP) regulates phosphorylation and function of the skeletal muscle ryanodine receptor. The Journal of Biological Chemistry. 2003; 278:24831–24836. [PubMed: 12709444]

Sag CM, Wadsack DP, Khabbazzadeh S, Abesser M, Grefe C, Neumann K, et al. Calcium/calmodulin-dependent protein kinase II contributes to cardiac arrhythmo-genesis in heart failure. Circulation. Heart failure. 2009; 2:664–675. [PubMed: 19919992]

Samso M, Feng W, Pessah IN, Allen PD. Corodinated movement of cytoplasmic and transmembrane domains of RyR1 upon gating. PLoS Biology. 2009; 7:e85. [PubMed: 19402748]

Sanchez G, Escobar M, Pedrozo Z, Macho P, Domenech R, Hartel S, et al. Exercise and tachycardia increase NADPH oxidase and ryanodine receptor-2 activity, possible role in cardioprotection. Cardiovascular Research. 2008; 77:380–386. [PubMed: 18006481]

Shin DW, Pan Z, Bandyopadhyay A, Bhat MB, Kim DH, Ma J. Ca^{2+}-dependent interaction between FKBP12 and calcineurin regulates activity of the Ca($^{2+}$) release channel in skeletal muscle. Biophysical Journal. 2002; 83:2539–2549. [PubMed: 12414688]

Souza JM, Peluffo G, Radi R. Protein tyrosine nitration-functional alteration or just a biomarker? Free Radical Biology and Medicine. 2008; 45:357–366. [PubMed: 18460345]

Stamler JS, Hausladen A. Oxidative modifications in nitrosative stress. Nature Structural Biology. 1998; 5:247–249.

Stamler JS, Meissner G. Physiology of nitric oxide in skeletal muscle. Physiological Reviews. 2001; 81:209–237. [PubMed: 11152758]

Stange M, Xu L, Balshaw D, Yamaguchi N, Meissner G. Characterization of recombinant skeletal muscle (Ser-2843) and cardiac muscle (Ser-2809) ryanodine receptor phosphorylation mutants. The Journal of Biological Chemistry. 2003; 278:51693–51702. [PubMed: 14532276]

Suko J, Drobny H, Hellmann G. Activation and inhibition of purified skeletal muscle calcium release channel by NO donors in single channel current recordings. Biochimica et Biophysica Acta. 1999; 1451:271–287. [PubMed: 10556582]

Suko J, Maurer-Fogy I, Plank B, Bertel O, Wyskovsky W, Hohenegger M, et al. Phosphorylation of serine 2843 in ryanodine receptor-calcium release channel of skeletal muscle by cAMP-, cGMP- and CaM-dependent protein kinase. Biochimica et Biophysica Acta. 1993; 1175:193–206. [PubMed: 8380342]

Sun J, Morgan M, Shen RF, Steenbergen C, Murphy E. Preconditioning results in S-nitrosylation of proteins involved in regulation of mitochondrial energetics and calcium transport. Circulation Research. 2007; 101:1155–1163. [PubMed: 17916778]

Sun J, Picht E, Ginsburg KS, Bers DM, Steenbergen C, Murphy E. Hypercontractile female hearts exhibit increased S-nitrosylation of the L-type Ca^{2+} channel alpha1 subunit and reduced ischemia/reperfusion injury. Circulation Research. 2006; 98:403–411. [PubMed: 16397145]

Sun J, Xin C, Eu JP, Stamler JS, Meissner G. Cysteine-3635 is responsible for skeletal muscle ryanodine receptor modulation by NO. Proceedings of the National Academy of Sciences of the United States of America. 2001; 98:11158–11162. [PubMed: 11562475]

Sun J, Xu L, Eu JP, Stamler JS, Meissner G. Classes of thiols that influence the activity of the skeletal muscle calcium release channel. The Journal of Biological Chemistry. 2001; 276:15625–15630. [PubMed: 11278999]

Sun J, Xu L, Eu JP, Stamler JS, Meissner G. Nitric oxide, NOC-12 and S-nitrosoglutathione modulate the skeletal muscle calcium release channel/ryanodine receptor by different mechanisms, An allosteric function for O_2 in S-nitrosylation of the channel. The Journal of Biological Chemistry. 2003; 278:8184–8189. [PubMed: 12509428]

Sun J, Yamaguchi N, Xu L, Eu JP, Stamler JS, Meissner G. Regulation of the cardiac muscle ryanodine receptor by O_2 tension and S-nitrosoglutathione. Biochemistry. 2008; 47:13985–13990. [PubMed: 19053230]

Takasago T, Imagawa T, Furukawa K, Ogurusu T, Shigekawa M. Regulation of the cardiac ryanodine receptor by protein kinase-dependent phosphorylation. Journal of Biochemistry (Tokyo). 1991; 109:163–170.

Takeshima H, Nishimura S, Matsumoto T, Ishida H, Kangawa K, Minamino N, et al. Primary structure and expression from complementary DNA of skeletal muscle ryanodine receptor. Nature. 1989; 339:439–445. [PubMed: 2725677]

Tang WH, Kravtsov GM, Sauert M, Tong XY, Hou XY, Wong TM, et al. Polyol pathway impairs the function of SERCA and RyR in ischemic-reperfused rat hearts by increasing oxidative modifications of these proteins. Journal of Molecular and Cellular Cardiology. 2010; 49:58–69. [PubMed: 20025885]

Tateishi H, Yano M, Mochizuki M, Suetomi T, Ono M, Xu X, et al. Defective domain-domain interactions within the ryanodine receptor as a critical cause of diastolic Ca^{2+} leak in failing hearts. Cardiovascular Research. 2009; 81:536–545. [PubMed: 18996969]

Terentyev D, Gyorke I, Belevych AE, Terentyeva R, Sridhar A, Nishijima Y, et al. Redox modification of ryanodine receptors contributes to sarcoplasmic reticulum Ca^{2+} leak in chronic heart failure. Circulation Research. 2008; 103:1466–1472. [PubMed: 19008475]

Tocchetti CG, Wang W, Froehlich JP, Huke S, Aon MA, Wilson GM, et al. Nitroxyl improves cellular heart function by directly enhancing cardiac sarcoplasmic reticulum Ca^{2+} cycling. Circulation Research. 2007; 100:96–104. [PubMed: 17138943]

Uehara A, Yasukochi M, Mejia-Alvarez R, Fill M, Imanaga I. Gating kinetics and ligand sensitivity modified by phosphorylation of cardiac ryanodine receptors. Pflugers Archiv. 2002; 444:202–212. [PubMed: 11976933]

Valdivia HH, Kaplan JH, Ellis-Davies GC, Lederer WJ. Rapid adaptation of cardiac ryanodine receptors, modulation by Mg^{2+} and phosphorylation. Science. 1995; 267:1997–2000. [PubMed: 7701323]

Voss AA, Lango J, Ernst-Russell M, Morin D, Pessah IN. Identification of hyperreactive cysteines within ryanodine receptor type 1 by mass spectrometry. The Journal of Biological Chemistry. 2004; 279:34514–34520. [PubMed: 15197184]

Wang P, Zweier JL. Measurement of nitric oxide and peroxynitrite generation in the postischemic heart. Evidence for peroxynitrite-mediated reperfusion injury. The Journal of Biological Chemistry. 1996; 271:29223–29230. [PubMed: 8910581]

Ward CW, Reiken S, Marks AR, Marty I, Vassort G, Lacampagne A. Defects in ryanodine receptor calcium release in skeletal muscle from post-myocardial infarct rats. The FASEB Journal. 2003; 17:1517–1519. [PubMed: 12824280]

Wehrens XH, Lehnart SE, Huang F, Vest JA, Reiken SR, Mohler PJ, et al. FKBP12.6 deficiency and defective calcium release channel (ryanodine receptor) function linked to exercise-induced sudden cardiac death. Cell. 2003; 113:829–840. [PubMed: 12837242]

Wehrens XH, Lehnart SE, Reiken SR, Deng SX, Vest JA, Cervantes D, et al. Protection from cardiac arrhythmia through ryanodine receptor-stabilizing protein calstabin2. Science. 2004; 304:292–296. [PubMed: 15073377]

Wehrens XH, Lehnart SE, Reiken SR, Marks AR. Ca^{2+}/calmodulin-dependent protein kinase II phosphorylation regulates the cardiac ryanodine receptor. Circulation Research. 2004; 94:e61–e70. [PubMed: 15016728]

Witcher DR, Kovacs RJ, Schulman H, Cefali DC, Jones LR. Unique phosphorylation site on the cardiac ryanodine receptor regulates calcium channel activity. The Journal of Biological Chemistry. 1991; 266:11144–11152. [PubMed: 1645727]

Xia R, Stangler T, Abramson JJ. Skeletal muscle ryanodine receptor is a redox sensor with a well defined redox potential that is sensitive to channel modulators. The Journal of Biological Chemistry. 2000; 275:36556–36561. [PubMed: 10952995]

Xia R, Webb JA, Gnall LL, Cutler K, Abramson JJ. Skeletal muscle sarcoplasmic reticulum contains a NADH-dependent oxidase that generates superoxide. The American Journal of Physiology. 2003; 285:C215–C221.

Xiao B, Jiang MT, Zhao M, Yang D, Sutherland C, Lai FA, et al. Characterization of a novel PKA phosphorylation site, serine-2030, reveals no PKA hyperphosphorylation of the cardiac ryanodine receptor in canine heart failure. Circulation Research. 2005; 96:847–855. [PubMed: 15790957]

Xiao B, Tian X, Xie W, Jones PP, Cai S, Wang X, et al. Functional consequence of protein kinase A-dependent phosphorylation of the cardiac ryanodine receptor, sensitization of store overload-induced Ca^{2+} release. The Journal of Biological Chemistry. 2007; 282:30256–30264. [PubMed: 17693412]

Xiao B, Zhong G, Obayashi M, Yang D, Chen K, Walsh MP, et al. Ser-2030, but not Ser-2808, is the major phosphorylation site in cardiac ryanodine receptors responding to protein kinase A activation upon beta-adrenergic stimulation in normal and failing hearts. The Biochemical Journal. 2006; 396:7–16. [PubMed: 16483256]

Xiao J, Tian X, Jones PP, Bolstad J, Kong H, Wang R, et al. Removal of FKBP12.6 does not alter the conductance and activation of the cardiac ryanodine receptor or the susceptibility to stress-induced ventricular arrhythmias. The Journal of Biological Chemistry. 2007; 282:34828–34838. [PubMed: 17921453]

Xin HB, Senbonmatsu T, Cheng DS, Wang YX, Copello JA, Ji GJ, et al. Oestrogen protects FKBP12.6 null mice from cardiac hypertrophy. Nature. 2002; 416:334–338. [PubMed: 11907581]

Xu KY, Huso DL, Dawson TM, Bredt DS, Becker LC. Nitric oxide synthase in cardiac sarcoplasmic reticulum. Proceedings of the National Academy of Sciences of the United States of America. 1999; 96:657–662. [PubMed: 9892689]

Xu L, Eu JP, Meissner G, Stamler JS. Activation of the cardiac calcium release channel (ryanodine receptor) by poly-S-nitrosylation. Science. 1998; 279:234–237. [PubMed: 9422697]

Yamaguchi N, Xin C, Meissner G. Identification of apocalmodulin and Ca^{2+}-calmodulin regulatory domain in skeletal muscle Ca^{2+} release channel, ryanodine receptor. The Journal of Biological Chemistry. 2001; 276:22579–22585. [PubMed: 11306590]

Yang D, Zhu WZ, Xiao B, Brochet DXP, Chen SRW, Lakatta EG, et al. Ca^{2+}/calmodulin kinase II-dependent phopshorylation of ryanodine receptors suppresses Ca^{2+} sparks and Ca^{2+} waves in cardiac myocytes. Circulation Research. 2007; 100:399–407. [PubMed: 17234969]

Yano M, Kobayashi S, Kohno M, Doi M, Tokuhisa T, Okuda S, et al. FKBP12.6-mediated stabilization of calcium-reelase channel (ryanodine receptor as a novel therapeutic strategey against heart failure. Circulation. 2003; 107:477–484. [PubMed: 12551874]

Yano M, Ono K, Ohkusa T, Suetsugu M, Kohno M, Hisaoka T, et al. Altered stoichiometry of FKBP12.6 versus ryanodine receptor as a cause of abnormal Ca^{2+} leak through ryanodine receptor in heart failure. Circulation. 2000; 102:2131–2136. [PubMed: 11044432]

Zahradnikova A, Minarovic I, Venema RC, Meszaros LG. Inactivation of the cardiac ryanodine receptor calcium release channel by nitric oxide. Cell Calcium. 1997; 22:447–454. [PubMed: 9502194]

Zalk R, Lehnart SE, Marks AR. Modulation of the ryanodine receptor and intracellular calcium. Ann. Rev. Biochem. 2007; 76:367–385. [PubMed: 17506640]

Zhang R, Khoo MS, Wu Y, Yang Y, Grueter CE, Ni G, et al. Calmodulin kinase II inhibition protects against structural heart disease. Natural Medicines. 2005; 11:409–417.

Zhang T, Maier LS, Dalton ND, Miyamoto S, Ross J Jr, Bers DM, et al. The deltaC isoform of CaMKII is activated in cardiac hypertrophy and induces dilated cardiomyopathy and heart failure. Circulation Research. 2003; 92:912–919. [PubMed: 12676814]

Zima AV, Blatter LA. Redox regulation of cardiac calcium channels and transporters. Cardiovascular Research. 2006; 71:310–321. [PubMed: 16581043]

Zissimopoulos S, Docrat N, Lai FA. Redox sensitivity of the ryanodine receptor interaction with FK506-binding protein. The Journal of Biological Chemistry. 2007; 282:6976–6983. [PubMed: 17200109]

Zorzato F, Fujii J, Otsu K, Phillips M, Green NM, Lai FA, et al. Molecular cloning of cDNA encoding human and rabbit forms of the Ca^{2+} release channel (ryanodine receptor) of skeletal muscle sarcoplasmic reticulum. The Journal of Biological Chemistry. 1990; 265:2244–2256. [PubMed: 2298749]

Zweier JL, Wang P, Kuppusamy P. Direct measurement of nitric oxide generation in the ischemic heart using electron paramagnetic resonance spectroscopy. The Journal of Biological Chemistry. 1995; 270:304–307. [PubMed: 7814391]