Mechanisms of Altered Excitation-Contraction Coupling in Canine Tachycardia-Induced Heart Failure, II

Model Studies

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Abstract—Ca$^{2+}$ transients measured in failing human ventricular myocytes exhibit reduced amplitude, slowed relaxation, and blunted frequency dependence. In the companion article (O’Rourke B, Kass DA, Tomaselli GF, Kääb S, Tunin R, Marbán E. Mechanisms of altered excitation-contraction coupling in canine tachycardia-induced heart failure, I: experimental studies. Circ. Res. 1999;84:562–570), O’Rourke et al show that Ca$^{2+}$ transients recorded in myocytes isolated from canine hearts subjected to the tachycardia pacing protocol exhibit similar responses. Analyses of protein levels in these failing hearts reveal that both SR Ca$^{2+}$ ATPase and phospholamban are decreased on average by 28% and that Na$^{+}$/Ca$^{2+}$ exchanger (NCX) protein is increased on average by 104%. In this article, we present a model of the canine midmyocardial ventricular action potential and Ca$^{2+}$ transient. The model is used to estimate the degree of functional upregulation and downregulation of NCX and SR Ca$^{2+}$ ATPase in heart failure using data obtained from 2 different experimental protocols. Model estimates of average SR Ca$^{2+}$ ATPase functional downregulation obtained using these experimental protocols are 49% and 62%. Model estimates of average NCX functional upregulation range are 38% and 75%. Simulation of voltage-clamp Ca$^{2+}$ transients indicates that such changes are sufficient to account for the reduced amplitude, altered shape, and slowed relaxation of Ca$^{2+}$ transients in the failing canine heart. Model analyses also suggest that altered expression of Ca$^{2+}$ handling proteins plays a significant role in prolongation of action potential duration in failing canine myocytes.

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Key Words: excitation-contraction coupling ■ heart failure ■ midmyocardial ventricular action potential ■ Ca$^{2+}$ transient

Recent studies using the canine tachycardia pacing-induced model of heart failure 1–8 demonstrate that changes in cellular electrophysiological and excitation-contraction (E-C) coupling processes are qualitatively similar to those observed in cells isolated from failing human heart. In human heart failure, $i_{K_1}$ current density measured at hyperpolarized membrane potentials is reduced by $\approx$ 50%,9,10 and density of the transient outward current $i_{Na_1}$ reduced by $\approx$ 75% in subepicardial11 and $\approx$ 40% in midmyocardial ventricular cells8 and is unchanged in subendocardial ventricular cells.11 The magnitude of $i_{K_1}$ is reduced by $\approx$ 40%, and that of $i_{Na_1}$ by $\approx$ 70% in failing canine midmyocardial cells.3 Expression of proteins involved in E-C coupling is also altered in human heart failure. Sarcoplasmic reticulum (SR) Ca$^{2+}$ ATPase mRNA levels,12–16 protein level,12,17,18 and uptake rate19 are reduced by $\approx$ 50% in end-stage heart failure. Na$^{+}$/Ca$^{2+}$ exchanger (NCX) mRNA levels are increased by $\approx$ 55% to 79%,12,20 and NCX protein levels increase 36% to 160%.12,20–22 Less information is available with regard to NCX function in heart failure. However, Reinecke et al22 reported an 89% increase in sodium-gradient–stimulated 45Ca$^{2+}$ uptake in human heart sarcolemmal vesicles.

As described in the preceding article by O’Rourke et al,23 alterations of intracellular Ca$^{2+}$ handling in failing canine midmyocardial ventricular myocytes parallel those observed in human. In particular, the time constant of Ca$^{2+}$ uptake in the absence of Na$^{+}$/Ca$^{2+}$ exchange is prolonged in failing cells (576±83 versus 282±30 ms in controls), suggesting a functional downregulation of the SERCA2a. This observation is consistent with Western blot analyses indicating that SR Ca$^{2+}$ ATPase protein levels are reduced in failing heart by 28%. Additionally, in the presence of cyclopiazonic acid (CPA, a blocker of the SR Ca$^{2+}$ ATPase pump), the time constant of Ca$^{2+}$ extrusion is larger in normal than failing cells (813±269 versus 599±48 ms). This observation is consistent with Western blot analyses indicating a 104%...
increase in the level of expression of the NCX in failing cells. Taken together, these results suggest that SR Ca\(^{2+}\) uptake is impaired and that Ca\(^{2+}\) extrusion via the NCX is enhanced in myocytes isolated from the failing canine heart in a way that is similar qualitatively to that seen in human patients.

In this article, we use the data of O'Rourke et al.\(^{23}\) to develop a computational model of the action potential and of intracellular Ca\(^{2+}\) handling in normal and failing canine ventricular myocytes using biophysically detailed descriptions of both sarclemmal currents and key components of E-C coupling. With the limits of individual alterations fixed using experimentally derived values, the model is used to quantify the extent to which each parameter (\(I_{\text{Ks}}, I_{\text{K1}}, \text{SR Ca}\(^{2+}\) ATPase, and NCX) contributes to the overall change in electrical and Ca\(^{2+}\) dynamics in heart failure. The results support the hypothesis that differences in expression of sarclemmal ion channels and Ca\(^{2+}\) handling proteins measured experimentally are sufficient to account for the altered action potential waveform and Ca\(^{2+}\) transient of the failing canine cardiomyocyte.

**Materials and Methods**

**Normal Canine Ventricular Cell Model**

Jafari et al.\(^{24}\) have presented a model of Ca\(^{2+}\) handling in the guinea pig ventricular myocyte that incorporates the following: (1) sarclemmal ion currents of the Luo-Rudy phase II ventricular cell model,\(^{25}\) (2) a state model of the L-type Ca\(^{2+}\) current in which Ca\(^{2+}\) -mediated inactivation occurs via the mechanism of mode switching,\(^{26}\) (3) calcium-induced calcium release from SR via ryanodine-sensitive calcium release (RyR) channels using a model adapted from that of Keizer and Levine,\(^{27}\) and (4) a restricted subspace located between the junctional SR (JSR) and T tubules into which both L-type Ca\(^{2+}\) and RyR channels empty. The model of the canine midmyocardial ventricular cell used in this study is derived from this guinea pig ventricular cell model. All dynamic equations, parameters, and initial conditions for this new model are given in the Appendix. The following modifications to the model of Jafari et al.\(^{24}\) have been made to better represent properties of canine midmyocardial ventricular cells.

\(I_{\text{Ks}}\)

Canine epicardial and midmyocardial ventricular cell action potentials exhibit a prominent notch in phase 1 of the action potential that results from the presence of 2 transient outward currents: a Ca\(^{2+}\) -dependent chloride (Cl–) current \(I_{\text{Cl}}\) and a Ca\(^{2+}\)-dependent current \(I_{\text{ClCa}}\).\(^{28,29}\) The Ca\(^{2+}\)-independent component \(I_{\text{Ks}}\) is modeled on the basis of the formulation of Campbell et al.\(^{31}\) for ferret ventricular cells. Peak \(I_{\text{Ks}}\) conductance \((G_{\text{Ks}})\) was adjusted to yield a linear plot of peak current density in response to 500-ms-duration voltage-clamp stimuli from a holding potential of −80 mV, with slope 0.3 pA/pF-mV and y-intercept 4.6 pA/PF. This agrees well with experimental measurements reported for canine \(I_{\text{Ks}}\) at 37°C by Liu et al.\(^{30}\) (see their Figure 10B: slope, 0.28 pA/PF-mV, and y-intercept, 5 pA/PF). Activation rate constants were scaled to yield a time to peak of ~8 ms at a clamp potential of +10 mV (see Figure 5B of Tseng and Hoffman).\(^{29}\) Inactivation rate constants were adjusted to yield a decay time constant of ~20 ms.\(^{29}\) The Ca\(^{2+}\)-dependent chloride (Cl–) current \(I_{\text{Cl}}\) was not incorporated in this model.

\(I_{\text{K1}}\)

The delayed rectifier current \(I_{\text{K1}}\) in both canine and guinea pig ventricular myocytes consists of rapid- and slow-activating components known as \(I_{\text{K1a}}\) and \(I_{\text{K1s}}\), respectively. Models of \(I_{\text{K1a}}\) and \(I_{\text{K1s}}\) in guinea pig ventricular cells have been developed.\(^{35}\) These models have been modified to approximate properties of corresponding currents measured in isolated canine midmyocardial ventricular cells. \(I_{\text{K1}}\) is described using a closed-open-state model in which forward \((K_{\text{f}})\) and backward \((K_{\text{b}})\) rate constants are exponential functions of voltage \((V)\) with the following form:

\[
K_{\text{f}}(V) = e^{-a(V-b)V},
\]

where \(a\) and \(b\) are parameters of the model.

Parameters of this model are fully constrained by knowledge of the time constant \(\tau(V)\), defined as

\[
\tau(V) = \frac{1}{K_{\text{f}}(V) + K_{\text{b}}(V)},
\]

at 2 voltages and by knowledge of the steady-state activation function. Activation was fit using a Boltzmann function determined by Liu and Antzelevitch\(^{31}\) (see their Figure 11). The time constant of activation at +5 mV was set to 100 ms,\(^{33}\) and the time constant of deactivation at −60 mV was set to 3000 ms,\(^{34}\) thereby constraining the rate constants \(K_{\text{f}}(V)\) and \(K_{\text{b}}(V)\). A fixed increment of 27 ms was added to Equation 2 to bound the time constant away from 0 at depolarized potentials. The maximum conductance \(G_{\text{Ka}}\) was adjusted to yield a tail current density of 0.2 pA/PF in response to a voltage-clamp step to +25 mV for 3.0 seconds, followed by a step to −35 mV for 1.0 seconds, as described by Gintant.\(^{35}\)

\(I_{\text{K1a}}\)

The slow-activating delayed rectifier current \(I_{\text{K1a}}\) is present in epicardial, midmyocardial, and endocardial canine ventricular cells. \(I_{\text{K1a}}\) is modeled as described in Zeng et al.\(^{32}\) with the exception that the steady-state activation function is fit using a Boltzmann function determined by Liu and Antzelevitch.\(^{33}\) The voltage-dependent time constant is also shifted by +40 mV in the depolarizing direction to fit the experimental data of Liu and Antzelevitch\(^{31}\) (see their Figure 13). Maximum conductance \((G_{\text{Ka}})\) is adjusted to yield a tail current density of 0.4 pA/PF in response to 3.0-second-duration voltage-clamp steps from the holding potential of −35 to +25 mV, followed by a return to the holding potential\(^{34}\) (see Figure 5). The Ca\(^{2+}\)-dependence of \(I_{\text{K1a}}\) described in the Luo-Rudy phase II guinea pig model is not included, as there are no experimental data constraining this dependence in canine ventricular cells.

\(I_{\text{K1s}}\)

\(I_{\text{K1s}}\) is fit using data measured at 22°C in isolated canine midmyocardial ventricular myocytes measured by Kääb et al.\(^{36}\) and scaled to 37°C. These data indicate that maximum outward \(I_{\text{K1s}}\) density is ~2.5 pA/PF at −60 mV\(^{36}\) (see Reference 5 , Figure 4B). These data also show that \(I_{\text{K1s}}\) density is nonnegligible at voltages within the plateau range of the canine action potential. For example, \(I_{\text{K1s}}\) density is 0.3 pA/PF at 0 mV, a value comparable with the density of \(I_{\text{K1a}}\) during the plateau phase of the action potential. The functional representation of \(I_{\text{K1s}}\) in the Luo-Rudy phase II model can therefore not be used, as it approaches 0 at plateau membrane potentials. An alternative formulation better approximating the canine data is presented in the Appendix.

\(I_{\text{Ca}}\)

The model of L-type Ca\(^{2+}\) current used is identical to the mode-switching model presented in Jafari et al.\(^{24}\) with 3 exceptions. First, the voltage dependence of the activation transition rates \(a(V)\) and \(b(V)\) and the inactivation variable \(y(V)\) are shifted by +10 mV in the depolarizing direction to position the peak L-type Ca\(^{2+}\) current in response to voltage-clamp stimuli at +5 mV, as measured experimentally.\(^{37}\) Second, the monotonous decreasing steady-state (voltage-dependent) inactivation function \(y(V)\) is modified to have an asymptotic value of 0.2 for large positive membrane potentials \(V\). This modification reproduces the slow component of Ca\(^{2+}\) current observed under voltage-clamp stimuli in canine ventricular cells.\(^{37}\) Finally, peak L-type Ca\(^{2+}\) current density is adjusted to a value of 2.5 pA/PF at a clamp voltage of +5 mV.

\(I_{\text{Ca}}\)

In the model of Jafari et al.\(^{24}\) Ca\(^{2+}\) uptake into network SR (NSR) is modeled using a Hill function with coefficient of 2.
rate is assumed to be 0, and Ca\(^{2+}\) leak from NSR to cytoplasm is assumed to be proportional to the gradient of NSR and cytosolic Ca\(^{2+}\) concentrations. Recently, Shannon et al\(^{16}\) have proposed the hypothesis that SR Ca\(^{2+}\) accumulation at rest is not limited by leak of Ca\(^{2+}\) from SR but rather is limited by a reverse component of SR Ca\(^{2+}\) ATPase pump current. They have proposed a new model of the SR Ca\(^{2+}\) ATPase pump that includes forward- and reverse-current components, each with its own binding constant and peak forward and reverse rates (denoted \(v_{\text{max}}\) and \(v_{\text{max}}\), respectively).\(^{25}\) The forward mode exhibits slight cooperativity, whereas the reverse mode is noncooperative. The relative magnitudes of forward- and reverse-current components determine whether SR load increases, is constant, or decreases during diastole. The model is presented in the Appendix.

**Failing Canine Ventrricular Cell Model**

Käib et al\(^{22}\) have shown that in the canine tachycardia pacing-induced model of heart failure, \(I_{\text{NI}}\) and \(I_{\text{Ki}}\) are downregulated on average by 66% and 32%, respectively, in terminal heart failure. Only the number of expressed channels is changed; the kinetic properties of \(I_{\text{Ni}}\) and gating behavior of \(I_{\text{Ki}}\) are unaltered. On the basis of these data, the effects of terminal heart failure are modeled by reducing the peak conductance of \(I_{\text{Na}}\) and \(I_{\text{K}}\) by the factors indicated above.

Downregulation of the SR Ca\(^{2+}\) ATPase is modeled by simultaneous scaling of both the forward and reverse maximum pump rates \(v_{\text{max}}\) and \(v_{\text{max}}\) by a scale factor, \(K_{\text{SR}}\). Uplregulation of the NCX is modeled by increasing a scale factor, \(K_{\text{NaCa}}\).

**Numerical Methods**

The dynamical equations in the Appendix are solved on a Silicon Graphics workstation using the Merson modified Runge-Kutta fourth-order adaptive step algorithm (No. 25, Reference 52), with a maximum step size of 100 microseconds and maximum error tolerance of 10\(^{-6}\). The error from all variables is normalized to ensure that each contributes equally to the calculation of global error, as described in Jafri et al\(^{24}\). Initial conditions listed in the Appendix are used in all calculations, unless noted otherwise. These initial conditions were computed in response to a periodic pulse train of frequency 1 Hz and were determined immediately before the 11th pulse. Action potentials are initiated using 0.1 \(\mu\text{A/pF}\) current injection for 500 microseconds.

The canine ventricular cell model is used to derive quantitative estimates of the NCX scale factor \(K_{\text{NaCa}}\) and the SR Ca\(^{2+}\) ATPase scale factor \(K_{\text{SR}}\) from experimental data by fitting model Ca\(^{2+}\) transient decay rates to those measured experimentally. To do this, a series of voltage-clamp stimuli (−97-mV holding potential, 3-mV step potential, and 200-ms duration) are applied at a frequency of 1 Hz. Ca\(^{2+}\) transient decay rate is estimated from response to the final voltage-clamp stimulus to assure that model SR Ca\(^{2+}\) concentrations have reached equilibrium values.

**Results**

**Action Potentials and Ca\(^{2+}\) Transients: Model Versus Experimental Results**

Figure 1 demonstrates the ability of the model to reconstruct action potentials and Ca\(^{2+}\) transients of both normal and failing canine midmyocardial ventricular myocytes. The solid and dotted lines in Figure 1A show experimental measurements of normal and failing action potentials, respectively. Model action potentials are shown in Figure 1C. In this figure, the solid line shows a normal action potential. The dashed line shows an action potential when \(I_{\text{Na}}\) is reduced by 66% of the normal values and \(I_{\text{K}}\) by 32% of the normal values (the average percentage reductions observed in terminal heart failure).\(^{3}\) The dotted line corresponds to these same reductions of \(I_{\text{Na}}\) and \(I_{\text{K}}\), in addition to a 62% reduction of the SR Ca\(^{2+}\) ATPase pump and a 75% increase of the NCX. These values are model-based estimates of the average percentage change in activity of these proteins determined using experimentally derived limits on their function, as described in the following sections.

The model data of Figure 1C show that downregulation of \(I_{\text{Na}}\) and \(I_{\text{K}}\) reduces the depth of the phase 1 notch. However, notch depth is larger in the experimental measurements from the failing myocyte (Figure 1A, dotted line) than is predicted by the model (Figure 1C, dashed line). This greater notch depth is due to the presence of the Ca\(^{2+}\)-dependent transient outward current \(I_{\text{tox}}\), which is not included in the model. The most significant change in model action potential duration (APD) occurs with upregulation of the NCX and downregulation of the SR Ca\(^{2+}\) ATPase (Figure 1C, dotted line). These 2 changes alone increase APDs at 90% repolarization (APD\(_{90}\)) by \(\approx 200\) ms.

Figure 1D illustrates model normal (solid line) and failing (dotted line) Ca\(^{2+}\) transients. Amplitude of the Ca\(^{2+}\) transient is reduced significantly in the heart failure model. Ca\(^{2+}\) transient shape is flattened, duration is prolonged, and relaxation is slowed. These changes are similar qualitatively to those seen in the experimental data of Figure 1B.

Figures 1E and 1F show L-type Ca\(^{2+}\) and Na\(^{+}\)/Ca\(^{2+}\) exchange currents for normal (solid lines) and failing (dotted lines) model cells. The reduction in peak magnitude of the L-type Ca\(^{2+}\) current seen in Figure 1E for the failing model cell results from downregulation of \(I_{\text{Na}}\), which reduces depth of the phase 1 notch and therefore driving force during onset of the L-type Ca\(^{2+}\) current. Figure 1E also shows that L-type Ca\(^{2+}\) current is increased during the later plateau phase of the action potential in failing model cells. The mechanism of this increase will be considered in subsequent sections. Figure 1F shows that Na\(^{+}\)/Ca\(^{2+}\) exchange operates in reverse mode, generating a net outward current during most of the plateau phase of the action potential. The magnitude of this outward current decreases during the plateau phase, and in the failing cell model the current becomes significantly smaller than the inward L-type Ca\(^{2+}\) current.

These simulations demonstrate the ability of the model to reproduce both normal and failing canine myocyte action potentials and Ca\(^{2+}\) transients. The following sections describe application of the model to estimation of the degree of functional change in the NCX and SR Ca\(^{2+}\) ATPase in control and failing myocytes. The approach is as follows: (1) the time constant of Ca\(^{2+}\) decay (\(\tau_{\text{Ca}}\)) measured with SR function blocked using CPA data is used to estimate the model Na\(^{+}\)/Ca\(^{2+}\) exchange scale factor \(K_{\text{NaCa}}\); (2) with \(K_{\text{NaCa}}\) fixed at this value, the model SR Ca\(^{2+}\) ATPase scale factor \(K_{\text{SR}}\) required to reproduce the \(\tau_{\text{Ca}}\) measured in physiological solutions is determined; (3) the SR Ca\(^{2+}\) ATPase reduction in heart failure is cross-checked independently by determining the model SR Ca\(^{2+}\) ATPase scale factor required to reproduce the \(\tau_{\text{Ca}}\) measured under Na\(^{+}\)-free conditions (0-Na data) with the model Na\(^{+}\)/Ca\(^{2+}\) exchange set to 0; and (4) the model Na\(^{+}\)/Ca\(^{2+}\) exchange scale factor is estimated independently from \(\tau_{\text{Ca}}\) in physiological solutions using the estimate of SR function determined in step 3.
Figure 1. Model vs experimental action potentials and Ca\(^{2+}\) transients. Each action potential and Ca\(^{2+}\) transient is in response to a 1-Hz pulse train, with responses measured in the steady state. A, Experimentally measured membrane potential as a function of time in normal (solid line) and failing (dotted line) canine myocytes. B, Experimentally measured cytosolic Ca\(^{2+}\) concentration (μmol/L) as a function of time for normal (solid line) and failing (dotted line) canine ventricular myocytes. C, Membrane potential as a function of time simulated using the normal canine myocyte model (solid line), the myocyte model with \(I_{\text{to1}}\) and \(I_{\text{K1}}\) downregulation (dashed line; downregulation by 66% and 32%, respectively), and the heart failure model (dotted line; downregulation of \(I_{\text{to1}}\) and \(I_{\text{K1}}\) as described previously, \(K_{\text{SR}}=0.38\) corresponding to 62% downregulation and \(K_{\text{NaCa}}=0.53\) corresponding to 75% upregulation). D, Cytosolic Ca\(^{2+}\) concentration (μmol/L) as a function of time simulated using the normal (solid line) and heart failure (dotted line) model, with parameters as described in panel A. E, L-type Ca\(^{2+}\) current as a function of time for the normal (solid line) and failing (dotted line) cell models. F, Na\(^{+}\)/Ca\(^{2+}\) exchange current as a function of time for the normal (solid line) and failing (dotted line) cell models.
downregulation of the NCX and SR Ca\(^{2+}\) ATPase in heart failure.

To estimate \(K_{NaCa}\) for \(K_{SR}\) was set to 0, 10 voltage-clamp steps (holding potential \(-97\) mV, step potential 3 mV, and duration 200 ms) were applied at a frequency of 1 Hz to ensure that Ca\(^{2+}\) levels in each model Ca\(^{2+}\) pool were equilibrated, and model \(\tau_{Ca}\) was measured by fitting an exponential function to the decay phase of the final Ca\(^{2+}\) transient. Figure 2A plots model \(\tau_{Ca}\) (ordinate, ms) as a function of \(K_{NaCa}\) (abscissa) with \(K_{SR}=0.0\) (open triangles). \(K_{NaCa}=0.30\) yields a \(\tau_{Ca}\) equal to the average value measured experimentally in normal myocytes in the presence of CPA (813±269 ms). One SD of experimental variability is accounted for by \(K_{NaCa}\) values in the interval (0.21, 0.48). This same curve shows that \(K_{NaCa}=0.53\) produces a \(\tau_{Ca}\) matching that measured in failing myocytes in the presence of CPA (599±248 ms). One SD of experimental variability is encompassed by \(K_{NaCa}\) values in the interval (0.48, 0.60). Assuming the normal value of \(K_{NaCa}\) to be 0.30, these data suggest a functional upregulation of the NCX in heart failure in the range of 60% to 100%, with average value \(\approx 75\%\).

Figure 2B plots model \(\tau_{Ca}\) (ordinate, ms) as a function of \(K_{SR}\) (abscissa). The curve marked with open circles plots this dependence when \(K_{NaCa}\) is constant at the normal value estimated above (\(K_{NaCa}=0.30\)). The experimental value of \(\tau_{Ca}\) measured in normal myocytes using physiological solutions is 219±36 ms. The maximum forward and reverse SR Ca\(^{2+}\) ATPase pump rates \(V_{max}\) and \(V_{max}\) given in Table 4 of the Appendix have been selected to yield a similar time constant when \(K_{SR}=1.0\). Measured variation about this value is accounted for by \(K_{SR}\) values in the interval (0.85, 1.15).

The experimental value of \(\tau_{Ca}\) measured in failing myocytes using physiological solutions is 292±23 ms. Dependence of model \(\tau_{Ca}\) on \(K_{SR}\) when \(K_{NaCa}\) is fixed at the value estimated for failing canine myocytes (0.53) is shown by the curve labeled with open squares in Figure 2B. \(K_{SR}=0.38\) yields a model \(\tau_{Ca}\) equal to that observed experimentally. The experimental deviation of \(\tau_{Ca}\) accounted for by \(K_{SR}\) values in the interval (0.26, 0.51). Assuming the average value of \(K_{SR}\) in normal cells to be 1.0, these data suggest a functional downregulation of the SR Ca\(^{2+}\) ATPase pump in heart failure in the range of 49% to 74%, with average value 62%.

**Estimation of NCX and SR Ca\(^{2+}\) ATPase Activity in Normal and Failing Myocytes:**

**CPA Experiments**

In the preceding article by O’Rourke et al, Ca\(^{2+}\) transients in response to voltage-clamp stimuli were measured in the presence and absence of CPA, a blocker of the SR Ca\(^{2+}\) ATPase pump. In the presence of CPA, Ca\(^{2+}\) transient decay rate (\(\tau_{Ca}\)) following termination of a depolarizing voltage step reflects the rate of extrusion of Ca\(^{2+}\) from the cytosol by the NCX (extrusion by the sarcolemmal Ca\(^{2+}\) ATPase is small). Estimates of the NCX pump current scale factor \(K_{NaCa}\) may therefore be obtained by setting the model value of \(K_{SR}\) to 0 and varying \(K_{NaCa}\) until model Ca\(^{2+}\) transient decay rates match those measured experimentally in the presence of CPA. \(K_{NaCa}\) may then be fixed at this value and \(K_{SR}\) varied until model Ca\(^{2+}\) transient decay rate matches that measured experimentally using physiological solutions. This procedure can be applied to data obtained from both normal and failing cells to assess the extent of functional upregulation and
decay rates match those measured experimentally using physiological solutions. This procedure can be applied to data obtained from both normal and failing cells to assess the extent of functional upregulation and downregulation of the NCX and SR Ca\(^{2+}\) ATPase in heart failure.

To mimic 0-Na conditions, \(K_{\text{NaCa}}\) was set equal to 0. \(K_{\text{SR}}\) was then varied, and the time constant for Ca\(^{2+}\) reuptake into SR was computed. Model \(\tau_{\text{Ca}}\) values are plotted as a function of \(K_{\text{SR}}\) in Figure 2B (open triangles). Experimentally measured values of this time constant are 282±30 ms in normal and 576±83 ms in failing canine ventricular cells studied under 0-Na conditions. A \(K_{\text{SR}}\) value of 1.0 accounts for \(\tau_{\text{Ca}}\) measured experimentally in normal cells (282±30 ms), and values in the interval (0.92, 1.07) account for the observed SD in these measurements. This estimate of the average \(K_{\text{SR}}\) value in normal myocytes based on block of the NCX agrees with that estimated using the CPA data. A \(K_{\text{SR}}\) value of 0.51 accounts for the average \(\tau_{\text{Ca}}\) measured experimentally in failing cells (576±83 ms), and \(K_{\text{SR}}\) values in the interval (0.46, 0.59) account for the SD. Assuming the normal \(K_{\text{SR}}\) value to be 1.0, these data suggest a functional downregulation of the SR Ca\(^{2+}\) ATPase pump in failing myocytes in the range of 41% to 54%, with average value 49%. This estimate of SR Ca\(^{2+}\) ATPase downregulation is qualitatively similar to that obtained using CPA.

Dependence of model \(\tau_{\text{Ca}}\) on \(K_{\text{NaCa}}\) when \(K_{\text{SR}}\) is fixed at the value estimated for normal canine myocytes (1.0) is shown by the curve labeled with open circles in Figure 2A. \(K_{\text{NaCa}}=0.22\) yields a model \(\tau_{\text{Ca}}\) equal to that observed experimentally using physiological solutions (219±36 ms). Experimental deviation of \(\tau_{\text{Ca}}\) is accounted for by \(K_{\text{NaCa}}\) values in the interval (0.13, 0.43). Dependence of model \(\tau_{\text{Ca}}\) on \(K_{\text{NaCa}}\) when \(K_{\text{SR}}\) is fixed at the value estimated for failing canine myocytes under 0-Na conditions (0.51) is shown by the curve labeled with open squares in Figure 2A. \(K_{\text{NaCa}}=0.35\) yields a model \(\tau_{\text{Ca}}\) equal to the average value observed experimentally using physiological solutions (292±23 ms). Experimental deviation of \(\tau_{\text{Ca}}\) is accounted for by \(K_{\text{NaCa}}\) values in the interval (0.26, 0.46). Assuming the normal value of \(K_{\text{NaCa}}\) to be 0.22, these data suggest a functional upregulation of the NCX in heart failure in the range of 18% to 109%, with average value 38%. This estimate of altered expression of NCX in heart failure has greater variability than that obtained previously using the CPA data but is consistent in that it also indicates increased expression.

**Parametric Dependence of Voltage-Clamp Ca\(^{2+}\) Transients on SR Ca\(^{2+}\) ATPase and NCX Levels**

The above analyses provide estimates of \(K_{\text{SR}}\) and \(K_{\text{NaCa}}\) in normal and failing myocytes. Results indicate functional downregulation of the SR Ca\(^{2+}\) ATPase pump and upregulation of the NCX in heart failure. The parametric dependence of model cytosolic Ca\(^{2+}\) transients on \(K_{\text{SR}}\) and \(K_{\text{NaCa}}\) is examined next.

Model cytosolic Ca\(^{2+}\) concentration (ordinate, \(\mu\text{mol/L}\)) versus time (abscissa, seconds) is shown in Figure 3A as \(K_{\text{SR}}\) is varied. In these simulations, \(K_{\text{NaCa}}\) is constant at the value estimated using CPA data from normal cells (\(K_{\text{NaCa}}=0.30\)). \(K_{\text{SR}}\) is varied from 1.0 to 0.0 in steps of 0.1. Ca\(^{2+}\) transients are in response to a 1-Hz voltage-clamp stimulus (holding potential −97 mV, step potential 3 mV, and duration 200 ms). Response to the final stimulus of 10 stimulus cycles is shown, with the time origin translated to 0 seconds. These data show that reduction of the model SR Ca\(^{2+}\) ATPase pump, simulating the effects of downregulation of this pump in heart failure, reduces the amplitude of the early peak of the Ca\(^{2+}\) transient (marked by the arrow). This early peak disappears as \(K_{\text{SR}}\) approaches 0. Figure 3B shows JSR Ca\(^{2+}\) levels for each of the responses in Figure 3A. Reduction of the early peak in the data of Figure 3A coincides with depletion of JSR Ca\(^{2+}\) at small values of \(K_{\text{SR}}\). Thus, the early peak in the model Ca\(^{2+}\) transient is generated by Ca\(^{2+}\) release from JSR, and the slow second peak, which is present even when JSR is depleted, results from influx of Ca\(^{2+}\) through sarcolemmal L-type Ca\(^{2+}\) channels and reverse-mode Na\(^{+}\)/Ca\(^{2+}\) exchange. As \(K_{\text{SR}}\) decreases, Ca\(^{2+}\) levels in JSR decrease, and the Ca\(^{2+}\) transient becomes reduced in peak amplitude. The Ca\(^{2+}\) transient exhibits a decrease, no change, or an increase of amplitude during the course of the voltage-clamp stimulus, depending on the value of \(K_{\text{SR}}\). Decay rate of the Ca\(^{2+}\) transient decreases.
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with decreasing $K_{SR}$ values, as shown in the data of Figure 3A, as well as Figure 2B (open circles).

Figure 4A shows model cytosolic Ca$^{2+}$ concentration (ordinate, $\mu$mol/L) versus time (abscissa, seconds) as $K_{NaCa}$ is varied in steps of 0.5 from 0.5 to 2.5. A plot for $K_{NaCa}=0.25$ is also shown. $K_{SR}$ is constant at the value estimated using CPA data from normal myocytes ($K_{SR}=1.0$). Voltage clamp steps from -97 mV to +3 mV with 200 ms duration are applied at a rate of 1.0 Hz. The final Ca$^{2+}$ transient in a sequence of 10 is displayed, with the time origin translated to 0 seconds. There are 3 effects of increased $K_{NaCa}$. These are (1) increased rate of Ca$^{2+}$ extrusion and lower diastolic Ca$^{2+}$ at the holding potential, (2) reduction in Ca$^{2+}$ transient amplitude in response to the +3 mV voltage-clamp step, and (3) “flattening” of the Ca$^{2+}$ transient during the voltage-clamp step. The increased Ca$^{2+}$ extrusion at the holding potential is a direct consequence of increased NCX activity when the exchanger is operating in the forward mode at the -97-mV holding potential, as shown in Figure 4B. This figure also shows that the NCX operates in reverse mode at the -97-mV potential. This produces a smaller Ca$^{2+}$ transient through reductions in SR Ca$^{2+}$ loading and therefore a smaller Ca$^{2+}$ release. The flattening of the Ca$^{2+}$ transient with increased $K_{NaCa}$ is a direct consequence of increased Ca$^{2+}$ influx during the voltage step, as shown in Figure 4B. Decreased $K_{NaCa}$ values also produce smaller Ca$^{2+}$ transient decay rates, as seen by the data of Figure 4A, as well as Figure 2A (open circles).

**Ca$^{2+}$ Transients in Response to Voltage-Clamp Stimuli: Model Versus Experimental Results**

Figure 5A shows model Ca$^{2+}$ transients in response to a 1-Hz voltage-clamp pulse train. These transients were computed using $K_{SR}$ and $K_{NaCa}$ parameter values determined from the experimental series in the presence and absence of CPA. The solid line is the normal model Ca$^{2+}$ transient ($K_{NaCa}=0.30$ and $K_{SR}=1.0$). The peak Ca$^{2+}$ level (480 nmol/mL) agrees well with the value measured experimentally in normal myocytes (450±75 nmol/mL). The dotted line is the model Ca$^{2+}$ transient computed using the average $K_{NaCa}$ (0.53) and $K_{SR}$ (0.38) values for failing myocytes. The remaining 2 Ca$^{2+}$ transients (dashed lines) correspond to $K_{NaCa}$ and $K_{SR}$ values selected at ±1 SD from the average for failing myocytes. The short dashed line represents parameter choices producing a high degree of SR unloading (large NCX activity, $K_{NaCa}=0.60$; small SR Ca$^{2+}$ ATPase activity, $K_{SR}=0.26$). The long-dashed line represents parameter choices that minimize SR unloading (small NCX activity, $K_{NaCa}=0.48$; large SR Ca$^{2+}$ ATPase activity, $K_{SR}=0.51$). These data show that as $K_{NaCa}$ is increased from a normal value of 0.30 (taking on values of 0.48, 0.53, and 0.60) and $K_{SR}$ is decreased from the normal value of 1.0 (taking on values 0.51, 0.38, and 0.26), Ca$^{2+}$ transient peak decreases monotonically from the normal value of 480 nmol/mL, taking on values of 300, 266, and 230 nmol/mL. These values agree well with the average experimental values measured in failing cells of 230±40 nmol/mL.

Figure 5B shows a Ca$^{2+}$ transient measured experimentally. The amplitude and waveform of the model predictions in Figure 5A are in close agreement with these experimental data.

Figure 5C shows a plot of the L-type Ca$^{2+}$ current during the Ca$^{2+}$ transients of Figure 5A. The parameter changes have relatively little effect on peak current, but increases in $K_{NaCa}$ or decreases in $K_{SR}$ produce a monotonic increase in the late component of the L-type Ca$^{2+}$ current. As shown in Figure 5D, these same parameter changes also produce monotonic decreases of the subspace Ca$^{2+}$ transient peak. Thus, the increase in the late component of L-type Ca$^{2+}$ current seen in Figure 5C results from a decrease in Ca$^{2+}$-mediated inactivation of this current due to reductions in magnitude of the subspace Ca$^{2+}$ transient, which is in turn a consequence of reduced SR Ca$^{2+}$ load. As can be appreciated by examining the magnitude of the change in L-type Ca$^{2+}$ current density with alterations in Ca$^{2+}$ handling, this late component of the L-type Ca$^{2+}$ current would be expected to play an important role in determining the action potential plateau. This suggests that in heart failure, alterations in the expression of Ca$^{2+}$ handling proteins that decrease SR Ca$^{2+}$ load and reduce the amplitude of the Ca$^{2+}$ transient may contribute substantially to prolongation of APD by reducing Ca$^{2+}$-mediated inactivation of the L-type Ca$^{2+}$ current.
In this article, we present a model of the canine midmyocardial action potential and Ca\(^{2+}\) transient. The model is used to estimate the magnitude of SR Ca\(^{2+}\) ATPase pump rates and NCX current in normal and failing myocytes using 2 methods. In the first method, model SR Ca\(^{2+}\) ATPase current is set to 0, and the NCX current is scaled to yield Ca\(^{2+}\) relaxation time constants in response to voltage-clamp stimuli matching those measured experimentally in normal and failing myocytes in the presence of CPA, a blocker of the SR Ca\(^{2+}\) ATPase. The extent of functional upregulation of the NCX in heart failure estimated using this approach is in the range of 60% to 100%, with average value 75%. Having constrained the model NCX current, model SR Ca\(^{2+}\) ATPase pump current is then estimated by matching the model Ca\(^{2+}\) relaxation rate to experimental data obtained in the absence of CPA. Comparison of model SR Ca\(^{2+}\) ATPase pump currents estimated for normal and failing myocytes suggests a functional downregulation in heart failure in the range of 49% to 74%, with average value 62%.

In the second method, model NCX current is set to 0, and the SR Ca\(^{2+}\) ATPase current is scaled to yield Ca\(^{2+}\) relaxation time constants matching those measured experimentally under 0-Na conditions. Functional downregulation of the SR Ca\(^{2+}\) ATPase current in heart failure estimated using this approach is in the range of 41% to 54%, with average value 49%. Having constrained the model SR Ca\(^{2+}\) ATPase current, NCX current is estimated by matching the model Ca\(^{2+}\) relaxation rate to experimental data obtained in control intracellular and extracellular sodium concentrations. Functional NCX upregulation in heart failure estimated using this approach is in the range of 18% to 109%, with average value 38%.

Analysis of protein levels in canine hearts subjected to the tachycardia pacing protocol reveal that both SR Ca\(^{2+}\) ATPase and phospholamban proteins are reduced on average by 28%\(^{23}\) and that NCX protein is increased on average by 104%.\(^{23}\) Both steady-state mRNA and expressed levels of E-C coupling proteins in failing human ventricular cells have
been measured. The majority of reports agree that there is a \( \approx 50\% \) reduction of: (1) mRNA encoding the SR Ca\(^{2+}\) ATPase pump,\(^{12-16} \) (2) expressed SR Ca\(^{2+}\) ATPase protein level,\(^{12,17,18} \) and (3) direct SR Ca\(^{2+}\) ATPase uptake rate during heart failure.\(^{19} \) There is a 55% to 79% increase in Na-Ca exchanger mRNA levels,\(^{12,20} \) a 36% to 160% increase in expressed protein levels,\(^{12,20-22} \) and an approximate factor of 2 increase in Na\(^+/\)Ca\(^{2+}\) exchange activity in human heart failure.\(^{22} \)

The model-based estimates of functional upregulation and downregulation of the NCX and SR Ca\(^{2+}\) ATPase pump reported here are consistent with these reports. Model estimates of average SR Ca\(^{2+}\) ATPase functional downregulation are 49% and 62%, depending on the estimation methods used. These values agree well with estimates of mRNA level, protein level, and SR Ca\(^{2+}\) ATPase uptake rate measured in human heart failure, but suggest a slightly larger degree of downregulation than indicated by measurements of protein level in canine tachycardia pacing-induced heart failure\(^{23} \). Model estimates of average NCX upregulation are 38% and 75%. These estimates agree well with measured increases in mRNA levels in human heart failure and are within the range of variability of measured NCX protein levels in human heart failure. However, the model estimates are slightly lower than is suggested by the increased protein levels measured in the failing canine heart.\(^{23} \)

Ca\(^{2+}\) transients measured in failing human and canine ventricular myocytes exhibit reduced amplitude and slowed relaxation.\(^{5,40-43} \) Model simulations of Ca\(^{2+}\) transients in response to voltage-clamp stimuli reported here demonstrate that the altered expression of the NCX and SR Ca\(^{2+}\) ATPase pump measured in failing canine myocytes is sufficient to account for these properties. Both changes contribute to reduced SR Ca\(^{2+}\) load and release and therefore reduced amplitude of the early Ca\(^{2+}\) transient peak (Figures 3A and 4A). The shape of the Ca\(^{2+}\) transient is also controlled by both NCX and SR Ca\(^{2+}\) ATPase levels. As the Ca\(^{2+}\) ATPase pump is downregulated (Figure 3A), the shape of the plateau portion of the voltage-clamp Ca\(^{2+}\) transient changes from negative to 0, then to positive slope. This change in slope is produced by a decrease in early Ca\(^{2+}\) release from JSR, which in turn increases the dependence of Ca\(^{2+}\) transient shape on Ca\(^{2+}\) entry through the L-type Ca\(^{2+}\) channel. Uptregulation of NCX also influences Ca\(^{2+}\) transient shape, tending to flatten the Ca\(^{2+}\) transient plateau by increasing reverse-mode Ca\(^{2+}\) entry at depolarized potentials (Figure 4A). The interplay between both of these factors accounts for the flattened Ca\(^{2+}\) transient shape seen in failing myocytes (Figure 1D, model; Figure 1B, experimental data).

Model Ca\(^{2+}\) transients in response to voltage-clamp stimuli exhibit a “knob” at the early peak of the transient (see Figure 3A, for example) that does not appear to be present in the experimental data. This knob disappears as the SR Ca\(^{2+}\) level becomes small (Figure 3A), indicating that the knob is dependent on SR Ca\(^{2+}\) release. The knob is likely an artifact of model construction. All SR Ca\(^{2+}\) release in this model occurs from a single functional unit, defined as a set of L-type Ca\(^{2+}\) channels, RyR channels, and the subspace within which they interact. Stern has referred to such models as common pool models.\(^{44} \) The knob reflects a large, single Ca\(^{2+}\) release event from this single functional unit. In contrast, real cardiac cells have a large number of functional units in which there is local control of calcium-induced calcium release. We have recently implemented a local control model of Ca\(^{2+}\) release consisting of an ensemble of functional units, in which each functional unit is defined as an L-type Ca\(^{2+}\) channel interacting with a small set of RyR channels through a diadic space. Both L-type Ca\(^{2+}\) channels and RyR channels are modeled stochastically using the channel models presented in Jafri et al.\(^{24} \) In such a model, the stochastic nature of RyR channel openings produces a variable latency of Ca\(^{2+}\) release in each functional unit. The Ca\(^{2+}\) transients computed using this model exhibit the property of gradedness and do not exhibit the knob seen in Figure 3A due to temporal smearing of Ca\(^{2+}\) release times.

A recent study has put forth the hypothesis that coupling between L-type Ca\(^{2+}\) channels and RyR channels may be altered in heart failure and that this altered coupling leads to a reduction in amplitude of the Ca\(^{2+}\) transient.\(^{45} \) The results presented here cannot refute this hypothesis. Indeed, structurally detailed models of RyR channel and L-type Ca\(^{2+}\) channel interactions in the diadic space predict a strong dependence of these interactions on geometric factors.\(^{46-48} \) However, the results reported here indicate that such an assumption is not necessary to account for reduced amplitude of Ca\(^{2+}\) transients in failing myocytes. Rather, these simulations indicate that the altered expression of Ca\(^{2+}\) handling proteins reported by several different groups in both failing human and canine myocytes could account for changes in Ca\(^{2+}\) transient amplitude and shape.

The data of Figure 1 demonstrate that downregulation of the outward repolarizing currents \( I_{\text{K1}} \) and \( I_{\text{Na}} \), together with altered expression of the NCX and SR Ca\(^{2+}\) ATPase pump, can account for differences in both action potential and Ca\(^{2+}\) transient shape in heart failure. However, the data of Figure 1C also indicate that downregulation of \( I_{\text{K1}} \) and \( I_{\text{Na}} \) at least to the extent measured on average in failing cells, has a small effect on APD. Instead, altered expression of Ca\(^{2+}\) handling proteins plays a significant role in APD prolongation.

It is not surprising that downregulation of model \( I_{\text{K1}} \) has only a modest impact on APD, as \( I_{\text{K1}} \) is primarily responsible for the terminal phase of repolarization. However, the finding that reduction of model \( I_{\text{Na}} \) has only a small effect on APD differs from the experimental results of Kääb et al\(^{45} \) in dog myocytes and of Beuckelmann et al\(^{45} \) in human cells. These experiments were performed using EGTA as an intracellular Ca\(^{2+}\) buffer. This buffering minimizes the modulatory effects of Ca\(^{2+}\) and thus enhances the relative influence of outward \( K \) currents on action potential characteristics. When effects of EGTA buffering are simulated in the model described in this article, block of \( I_{\text{Na}} \) has a greater influence on APD. An example is shown in Figure 6. The Ca\(^{2+}\) buffering effects of EGTA were modeled using the fast buffering approximation developed by Wagner and Keizer,\(^{49} \) with EGTA = 10 mmol/L and the dissociation constant \( K_{\text{diss}} = 0.15 \) mmol/L. Block of \( I_{\text{Na}} \) by 95% increases APD\(_{90}\) by 73 ms, or \( \approx 25\% \) of the control value. These results again emphasize the important modula-
tory role of Ca\(^{2+}\) on action potential characteristics in the canine myocyte.

It is also possible that 4-AP block of K currents other than I\(_{\text{so}}\) occurred in the Kääb et al\(^5\) experiments, but that such effects were not resolvable. Steady-state current-voltage relations were measured in the presence and absence of 4-AP to assess whether or not this was the case. Data are shown in Figure 10C of Kääb et al\(^5\) and indicate that experimental variability in steady-state current at 0 mV (a potential near that of the action potential plateau) is roughly \pm 1.0 pA/pF. The sum of model outward currents \(I_{\text{to}}, I_{\text{kr}}, I_{\text{kr}}\), and \(I_{\text{cl}}\) during the plateau is comparable with the magnitude of this variability in the experimental measurements (\(\approx 1.0\) pA/pF). Genetic approaches for selective suppression of \(I_{\text{so}}\)\(^5, 51\) may turn out to be more useful than pharmacological approaches in determining the influence of this current on APD.

The model predicts that one important mechanism of APD prolongation in heart failure is that shown in Figures 1 and 5. Under conditions of reduced SR Ca\(^{2+}\) release, there is less Ca\(^{2+}\)-mediated inactivation of the L-type Ca\(^{2+}\) current. The resulting increase of inward current, as shown for voltage-clamp stimuli in Figure 5C and for action potentials in Figure 1E, helps to maintain and prolong the plateau phase of the action potential. Investigation into the relative contribution of the various Ca\(^{2+}\)-regulatory mechanisms and Ca\(^{2+}\)-dependent membrane currents in determining the action potential shape and duration is an important area for future experimental and modeling studies.

**Figure 6.** Membrane potential as a function of time for the normal model (solid line) and for the model with a 95% reduction in magnitude of \(I_{\text{so}}\) (dotted line). Simulations are done in the presence of EGTA (10 mmol/L; \(K_{\text{m}}=0.15\) μmol/L), using the fast buffering approximation of Wagner and Keizer.\(^{39}\) Response to fifth stimulus at a cycle length of 1200 ms is shown.

**Appendix**

**Standard Units (Unless Otherwise Noted) Used in the Following Set of Equations**

<table>
<thead>
<tr>
<th>Quantity</th>
<th>Units</th>
</tr>
</thead>
<tbody>
<tr>
<td>Membrane potential</td>
<td>mV</td>
</tr>
<tr>
<td>Membrane current</td>
<td>μA/μF(^{-1})</td>
</tr>
<tr>
<td>Membrane conductance</td>
<td>mS/μF(^{-1})</td>
</tr>
<tr>
<td>Ionic flux</td>
<td>mM/μs(^{-1})</td>
</tr>
<tr>
<td>Concentration</td>
<td>mM</td>
</tr>
<tr>
<td>Time constant</td>
<td>ms</td>
</tr>
<tr>
<td>Rate constant</td>
<td>ms(^{-1})</td>
</tr>
</tbody>
</table>

**Membrane Currents**

**Na\(^+\) Current \(I_{\text{Na}}\)**

(A.1) \[ I_{\text{Na}} = G_{\text{Na}} m^3 h_j (V - E_{\text{Na}}) \]

(A.2) \[ E_{\text{Na}} = \frac{RT}{F} \ln \left( \frac{[\text{Na}^+]_o}{[\text{Na}^+]_i} \right) \]

(A.3) \[ \frac{dm}{dt} = \alpha_m (1 - m) - \beta_m m \]

(A.4) \[ \frac{dh}{dt} = \alpha_h (1 - h) - \beta_h h \]

(A.5) \[ \frac{dj}{dt} = \alpha_j (1 - j) - \beta_j j \]

(A.6) \[ \alpha_m = 0.32 \frac{V + 47.13}{1 - e^{-0.1(1 + 47.13)^{0.35}}} \]

(A.7) \[ \beta_m = 0.08 e^{-\frac{V}{17}} \]

For \(V \leq -40\) mV, \(\alpha_h = 0\)

(A.8) \[ \alpha_h = 0 \]

(A.9) \[ \beta_h = \frac{1}{0.13(1 + e^{V - 10.089/11.3})} \]

(A.10) \[ \alpha_j = 0 \]

(A.11) \[ \beta_j = 0.3 \frac{e^{-2.535 \times 10^{-7} V}}{1 + e^{-0.119 - 7.32}} \]

For \(V < -40\) mV, \(\alpha_h = 0.135 e^{(80 + 9V)^{-6.8}} \)

(A.12) \[ \beta_h = 3.56 e^{-0.079V} + 3.1 \times 10^6 e^{-0.35V} \]

(A.13) \[ \alpha_i = (-127140 e^{0.2541V} - 3.474 \]

\[ \times 10^{-5} e^{-0.0453 V}) (V + 37.78)^{1.5} (1 + e^{6.311 V + 9.237}) \]

(A.14) \[ \beta_i = 0.121 e^{-0.01052V} \]

\[ \frac{e^{-0.117V} + 9.40.143}{1 + e^{-0.117V} + 9.40.143} \]

**Rapid-Activating Delayed Rectifier K\(^+\) Current \(I_{\text{Kr}}\)**

(A.15) \[ I_{\text{Kr}} = G_{\text{Kr}} f([\text{K}^+]_o) R(V) X_{\text{Kr}} (V - E_{\text{Kr}}) \]
\[ E_K = \frac{RT}{F} \ln \left( \frac{[K^+]_o}{[K^+]_1} \right) \]

\[ R(V) = \frac{1}{1 + 1.4945e^{0.1097V}} \]

\[ f(K^+) = \sqrt{([K^+]_1)^4} \]

\[ \frac{dX_{K_1}}{dt} = K_{12} (1 - X_{K_1}) - K_{21} X_{K_1} \]

\[ K_{12} = e^{-5.945 + 0.1691V} \]

\[ K_{21} = e^{-7.677 - 0.0126V} \]

**Slow-Activating Delayed Rectifier K⁺ Current \( I_{K_s} \)**

\[ I_{K_s} = \frac{\tilde{G}_{K_s} X_{K_s}(V - E_{K_s})}{1 + e^{(V - 14.5)/24.931}} \]

\[ \tau_{K_s} = \frac{0.0000719(V - 10)}{1 - e^{-104(V - 10)/24.931}} \times \frac{0.000131(V - 10)}{1 - e^{-104(V - 10)/24.931}} \]

**Transient Outward K⁺ Current \( I_{To} \)**

\[ I_{To} = \tilde{G}_{To} X_{To} Y_{To}(V - E_K) \]

\[ \frac{dX_{To}}{dt} = \alpha_{X_to}(1 - X_{To}) - \beta_{X_to} X_{To} \]

\[ \frac{dY_{To}}{dt} = \alpha_{Y_to}(1 - Y_{To}) - \beta_{Y_to} Y_{To} \]

\[ \alpha_{X_to} = 0.04516e^{0.0357TV} \]

\[ \beta_{X_to} = 0.0989e^{-0.0625TV} \]

\[ \alpha_{Y_to} = 0.005415e^{-15V + 3.5} \times 10^{-5} \]

\[ \beta_{Y_to} = 0.005415e^{-15V + 3.5} \times 10^{-5} \]

**Time-Independent K⁺ Current \( I_{Ki} \)**

\[ I_{Ki} = \tilde{G}_{Ki} K_i(V) \left( \frac{[K^+]_o + K_{mk1}}{[K^+]_o} \right) (V - E_K) \]

\[ K_i(V) = \frac{1}{2 + e^{(V - 10)/9}} \]

**Plateau K⁺ Current \( I_{Kp} \)**

\[ I_{Kp} = \tilde{G}_{Kp} K_p(V)(V - E_K) \]

\[ K_p(V) = \frac{1}{1 + e^{17.488 - 905.95}} \]

**NCX Current \( I_{Na,Ca} \)**

\[ I_{Na,Ca} = k_{Na,Ca}[Na^+]_o [Ca^{2+}]_i + [Ca^{2+}]_o + [Na^+]_i + 1 + k_{Na,Ca}e^{(V - 10)/30} \times \]

\[ (e^{30/RT} \times [Na^+][Ca^{2+}] - e^{(V - 10)/30}RT [Na^+][Ca^{2+}]) \]

**Na⁺-K⁺ Pump Current \( I_{Na,K} \)**

\[ I_{Na,K} = \tilde{I}_{Na,K} \frac{1}{1 + e^{(V - 10)/10}} \times \frac{[K^+]_o}{[Na^+]_o} + 1 \]

\[ f_{Na,K} = 0.1245e^{0.01097V} + 0.0365e^{0.01097V} \]

**Sarcosomal Ca²⁺ Pump Current \( I_{p(Ca)} \)**

\[ I_{p(Ca)} = \tilde{I}_{p(Ca)} \frac{[Ca^{2+}]_o}{K_{mp(Ca)} + [Ca^{2+}]_o} \]

**Ca²⁺ Background Current \( I_{Ca,b} \)**

\[ I_{Ca,b} = \tilde{G}_{Ca,b}(V - E_{Ca}) \]

\[ E_{Ca} = \frac{RT}{2F} \ln \left( \frac{[Ca^{2+}]_o}{[Ca^{2+}]_o} \right) \]

**Na⁺ Background Current \( I_{Na,b} \)**

\[ I_{Na,b} = \tilde{G}_{Na,b}(V - E_{Na}) \]

**Membrane Potential**

\[ \frac{dV}{dt} = -I_{Na} + I_{Ca} + I_{Na,K} + I_{Ca,b} + I_{Ca,C} + I_{Kp} + I_{Na,Ca} + I_{Na,K} + I_{Na,Ca} + I_{p(Ca)} + I_{Ca,b} + I_{Na,b} \]

**Ca²⁺ Handling Mechanisms**

**L-Type Ca²⁺ Current \( I_{Ca} \)**

\[ \alpha = 0.4e^{(V + 2.1)/10} \]

\[ \beta = 0.05e^{-(V + 2.1)/13} \]

\[ \alpha' = \alpha \]

\[ \beta' = \frac{\beta}{b} \]

\[ \gamma = 0.10375[Ca^{2+}]_o \]

\[ \frac{dC_i}{dt} = \beta C_i + \omega C_{Ca,i} - (\alpha + \gamma)C_0 \]

\[ \frac{dC_1}{dt} = 4\alpha C_i + 2\beta C_2 + \omega C_{Ca,1} \]

\[ \frac{dC_2}{dt} = 3\alpha C_1 + 3\beta C_3 + \omega C_{Ca,2} \]

\[ - (\beta + 3\alpha + \gamma)C_1 \]

\[ - (2\beta + 2\alpha + \gamma)C_2 \]
E-C Coupling in Heart Failure II

\[
\frac{dC_1}{dt} = 2\alpha C_1 + 4\beta C_4 + \frac{\omega}{b} C_{C3} - \left(3\beta + \alpha + \gamma C_1\right)
\]

\[
\frac{dC_4}{dt} = \alpha C_4 + g O + \frac{\omega}{b} C_{C4} - \left(4\beta + \frac{f + \gamma a}{b} C_4\right)
\]

\[
\frac{dO}{dt} = f C_4 - g O
\]

\[
\frac{dC_{C4}}{dt} = \beta C_{C4} + \gamma C_i - (4\alpha + \alpha C_{C4} + \gamma C_1
\]

\[
\frac{dC_{C2}}{dt} = 2\alpha C_{C2} + 4\beta C_{C4} + \gamma a C_i
\]

\[
\frac{dC_{C3}}{dt} = 3\alpha C_{C3} + 3\beta C_{C3} + \gamma a C_2
\]

\[
\frac{dC_{C4}}{dt} = \alpha C_{C4} + g O_{C4} + \gamma a C_4
\]

\[
\frac{dO_{C4}}{dt} = f C_{C4} - g O_{C4}
\]

\[
\frac{P_{C5} 4VF^2}{C_{Ca}} 0.001e^{\frac{V}{FRT}} - 0.341[Ca^{2+}]_i
\]

\[
I_{Ca} = I_{Ca,y}[O + O_{Ca}]
\]

\[
I_{Ca,y} = \frac{P_{Ca}'}{C_{Ca}}[O + O_{Ca}]
\]

\[
Ve^2 [K^+]_i e^{\frac{V}{FRT}} - [K^+]_i
\]

\[
P_{Ca}' = \frac{\tilde{P}_{Ca}}{1 + \frac{I_{Ca}}{I_{Ca,0}}}
\]

\[
\frac{dy}{dt} = \frac{y_s - y}{\tau_y}
\]

\[
y_s = \frac{0.8}{1 + e^{\frac{V}{FRT_{SST}}}} + 0.2
\]

\[
\tau_y = 20 + \frac{600}{1 + e^{\frac{V}{FRT_{SST}}}}
\]

RyR Channel (Keizer and Levine)\textsuperscript{27}

\[
\frac{dC_{Ca}}{dt} = -k_s [Ca^{2+}]_i P_{Ca} + k_s P_{o2}
\]

\[
\frac{dP_{o2}}{dt} = k_o [Ca^{2+}]_i P_{Ca} - k_{o2} P_{o2} - k_{o2} P_{Ca}
\]

\[
\frac{dP_{o2}}{dt} = -k_{o2} P_{Ca} - k_{o2} P_{o2}
\]

\[
\frac{dP_{o1}}{dt} = k_o [Ca^{2+}]_i P_{Ca} + k_{o1} P_{o1} + k_{o1} P_{Ca}
\]

\[
\frac{dP_{o1}}{dt} = k_o [Ca^{2+}]_i P_{Ca} + k_{o1} P_{o1} + k_{o1} P_{Ca}
\]

\[
J_{Ca} = v_i (P_{o1} + P_{o2}) [Ca^{2+}]_i [Ca^{2+}]_o
\]

**SERCA2a Pump (Shannon et al)\textsuperscript{18}**

\[
f_s = [Ca^{2+}]_K / [Ca^{2+}]_SR
\]

\[
r_s = [Ca^{2+}]_SR / [Ca^{2+}]_K
\]

\[
J_{up} = k_{up} [P_{o1} + P_{o2}] [Ca^{2+}]_i - [Ca^{2+}]_o
\]

**Intracellular Ca\textsuperscript{2+} Fluxes**

\[
J_{u} = \frac{[Ca^{2+}]_SR - [Ca^{2+}]_i}{\tau_{u}}
\]

\[
J_{sdr} = \frac{[Ca^{2+}]_L - [Ca^{2+}]_i}{\tau_{sdr}}
\]

\[
J_{up} = \frac{d[HTRPNCa]}{dt} + \frac{d[LTRPNCa]}{dt}
\]

\[
\frac{d[HTRPNCa]}{dt} = k_{up} [Ca^{2+}]_i ([HTRPNCa]_i - [HTRPNCa])
\]

\[
\frac{d[LTRPNCa]}{dt} = k_{up} [Ca^{2+}]_i ([HTRPNCa]_i - [LTRPNCa])
\]

**Intracellular Ion Concentrations**

\[
\frac{d}[Na^+]_i}{dt} = -(I_{Na} + I_{Na,k} + 3I_{Na,c} + 3I_{Na,k} + \frac{A_{Na,Ca}}{V_{m,F}})
\]

\[
\frac{d}[K^+]_i}{dt} = -(I_{K_r} + I_{K_e} + I_{Na} + I_{K_p} + I_{Ca,k})
\]

\[
\frac{d}[Ca^{2+}]_i}{dt} = \frac{A_{Ca,Ca}}{V_{m,F}}
\]

\[
\frac{d}[Ca^{2+}]_i}{dt} = \beta_{s} \frac{J_{ss} V_{ss}}{V_{m}} - (I_{Ca} \frac{A_{Na,Ca}}{2V_{m,F}})
\]
\[
\frac{d[Ca^{2+}]_{\text{JSR}}}{dt} = \beta_{\text{JSR}}(J_u - J_{\text{st}})
\]
\[
\frac{d[Ca^{2+}]_{\text{NSR}}}{dt} = J_{\text{myo}}V_{\text{myo}} - J_{\text{p}}V_{\text{NSR}}
\]

### Tables

#### TABLE 1. Cell Geometry Parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Definition</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>(C_{\text{sc}})</td>
<td>Specific membrane capacity</td>
<td>1.00 (\mu)F cm(^{-2})</td>
</tr>
<tr>
<td>(A_{\text{cap}})</td>
<td>Capacitive membrane area</td>
<td>(1.534 \times 10^{-2}) cm(^2)</td>
</tr>
<tr>
<td>(V_{\text{JSR}})</td>
<td>JSR volume</td>
<td>(25.84 \times 10^{-6}) (\mu)L</td>
</tr>
<tr>
<td>(V_{\text{NSR}})</td>
<td>NSR volume</td>
<td>(0.16 \times 10^{-6}) (\mu)L</td>
</tr>
<tr>
<td>(V_{\text{SS}})</td>
<td>Subspace volume</td>
<td>(1.2 \times 10^{-9}) (\mu)L</td>
</tr>
</tbody>
</table>

#### TABLE 2. Standard Ionic Concentrations

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Definition</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>([K^+]_o)</td>
<td>Extracellular K(^+) concentration</td>
<td>4.0 mmol/L</td>
</tr>
<tr>
<td>([Na^+]_o)</td>
<td>Extracellular Na(^-) concentration</td>
<td>138.0 mmol/L</td>
</tr>
<tr>
<td>([Ca^{2+}]_o)</td>
<td>Extracellular Ca(^{2+}) concentration</td>
<td>2.0 mmol/L</td>
</tr>
</tbody>
</table>

#### TABLE 3. Membrane Current Parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Definition</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>(F)</td>
<td>Faraday constant</td>
<td>96.5 (\text{coulomb mmol}^{-1})</td>
</tr>
<tr>
<td>(T)</td>
<td>Absolute temperature</td>
<td>310 K</td>
</tr>
<tr>
<td>(R)</td>
<td>Ideal gas constant</td>
<td>8.314 (\text{J} \text{mol}^{-1} \text{K}^{-1})</td>
</tr>
<tr>
<td>(g_{\text{IK}})</td>
<td>Peak (I_{K}) conductance</td>
<td>0.0034 (\text{mS} \mu\text{F}^{-1})</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Peak (I_{K1}) conductance</td>
<td>0.00271 (\text{mS} \mu\text{F}^{-1})</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Peak (I_{K1}) conductance</td>
<td>0.23815 (\text{mS} \mu\text{F}^{-1})</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Peak (I_{K1}) conductance</td>
<td>2.8 (\text{mS} \mu\text{F}^{-1})</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Peak (I_{Kp}) conductance</td>
<td>0.002216 (\text{mS} \mu\text{F}^{-1})</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Peak (I_{K1}) conductance</td>
<td>0.128 (\text{mS} \mu\text{F}^{-1})</td>
</tr>
<tr>
<td>(k_{\text{sat}})</td>
<td>Na(^+)-Ca(^{2+}) exchange saturation factor at negative potentials</td>
<td>0.30 (\text{mS} \mu\text{F}^{-1})</td>
</tr>
<tr>
<td>(h)</td>
<td>Controls voltage dependence of Na(^+)-Ca(^{2+}) exchange</td>
<td>0.35</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Maximum Na(^+)-K(^-) pump current</td>
<td>0.963 (\mu\text{A}\mu\text{F}^{-1})</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Na(^+) half-saturation constant for Na(^+)-K(^-) pump</td>
<td>10.0 mmol/L</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Na(^+) half-saturation constant for Na(^+)-K(^-) pump</td>
<td>1.5 mmol/L</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Maximum sarcolemmal Ca(^{2+}) pump current</td>
<td>0.05 (\mu\text{A}\mu\text{F}^{-1})</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Half-saturation constant for sarcolemmal Ca(^{2+}) pump</td>
<td>0.00005 (\text{mmol} \text{L}^{-1})</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Maximum background Ca(^{2+}) conductance</td>
<td>0.0003842 (\text{mS} \mu\text{F}^{-1})</td>
</tr>
<tr>
<td>(g_{\text{IK1}})</td>
<td>Maximum background Na(^+) current conductance</td>
<td>0.031 (\text{mS} \mu\text{F}^{-1})</td>
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</tbody>
</table>
### Table 4. SR Parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Definition</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>( v )</td>
<td>Maximum ( \mathrm{Ca}^{2+} ) flux</td>
<td>1.8 ms(^{-1} )</td>
</tr>
<tr>
<td>( K_{f} )</td>
<td>Forward half-saturation constant for ( \mathrm{Ca}^{2+} ) ATPase</td>
<td>0.168 \times 10^{-3} \text{ mmol/L}</td>
</tr>
<tr>
<td>( K_{b} )</td>
<td>Backward half-saturation constant for ( \mathrm{Ca}^{2+} ) ATPase</td>
<td>3.29 \text{ mmol/L}</td>
</tr>
<tr>
<td>( K_{\text{Sr}} )</td>
<td>Scaling factor for ( \mathrm{Ca}^{2+} ) ATPase</td>
<td>1.0</td>
</tr>
<tr>
<td>( N_{f} )</td>
<td>Forward cooperativity constant for ( \mathrm{Ca}^{2+} ) ATPase</td>
<td>1.2</td>
</tr>
<tr>
<td>( N_{b} )</td>
<td>Reverse cooperativity constant for ( \mathrm{Ca}^{2+} ) ATPase</td>
<td>1.0</td>
</tr>
<tr>
<td>( v_{\text{cat}} )</td>
<td>( \mathrm{Ca}^{2+} ) ATPase forward rate parameter</td>
<td>0.813 \times 10^{-4} \text{ mmol/L ms}^{-1}</td>
</tr>
<tr>
<td>( v_{\text{off}} )</td>
<td>( \mathrm{Ca}^{2+} ) ATPase reverse rate parameter</td>
<td>0.318 \times 10^{-3} \text{ mmol/L ms}^{-1}</td>
</tr>
<tr>
<td>( t_{\text{tr}} )</td>
<td>Time constant for transfer from NSR to JSR</td>
<td>0.5747 ms</td>
</tr>
<tr>
<td>( t_{\text{xfer}} )</td>
<td>Time constant from subspace to myoplasm</td>
<td>26.7 ms</td>
</tr>
<tr>
<td>( k_{+} )</td>
<td>RyR ( P_{i} ) – ( P_{o} ) rate constant</td>
<td>12.15 \times 10^{9} \text{ mmol/L ms}^{-1}</td>
</tr>
<tr>
<td>( k_{-} )</td>
<td>RyR ( P_{o} ) – ( P_{i} ) rate constant</td>
<td>0.576 ms^{-1}</td>
</tr>
<tr>
<td>( k_{c} )</td>
<td>RyR ( P_{o} ) – ( P_{o} ) rate constant</td>
<td>4.05 \times 10^{6} \text{ mmol/L ms}^{-1}</td>
</tr>
<tr>
<td>( k_{c} )</td>
<td>RyR ( P_{i} ) – ( P_{i} ) rate constant</td>
<td>1.930 ms^{-1}</td>
</tr>
<tr>
<td>( k_{c} )</td>
<td>RyR ( P_{o} ) – ( P_{i} ) rate constant</td>
<td>0.100 ms^{-1}</td>
</tr>
<tr>
<td>( k_{c} )</td>
<td>RyR ( P_{o} ) – ( P_{o} ) rate constant</td>
<td>0.0008 ms^{-1}</td>
</tr>
<tr>
<td>( n )</td>
<td>RyR ( \mathrm{Ca}^{2+} ) cooperativity parameter ( P_{i} ) – ( P_{o} )</td>
<td>4</td>
</tr>
<tr>
<td>( m )</td>
<td>RyR ( \mathrm{Ca}^{2+} ) cooperativity parameter ( P_{i} ) – ( P_{o} )</td>
<td>3</td>
</tr>
</tbody>
</table>

### Table 5. L-Type \( \mathrm{Ca}^{2+} \) Channel Parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Definition</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>( f )</td>
<td>Transition rate into open state</td>
<td>0.3 ms(^{-1} )</td>
</tr>
<tr>
<td>( g )</td>
<td>Transition rate out of open state</td>
<td>2.0 ms(^{-1} )</td>
</tr>
<tr>
<td>( f' )</td>
<td>Transition rate into open state for mode Ca</td>
<td>0.005 ms(^{-1} )</td>
</tr>
<tr>
<td>( g' )</td>
<td>Transition rate out of open state for mode Ca</td>
<td>7.0 ms(^{-1} )</td>
</tr>
<tr>
<td>( b )</td>
<td>Mode transition parameter</td>
<td>2.0</td>
</tr>
<tr>
<td>( a )</td>
<td>Mode transition parameter</td>
<td>2.0</td>
</tr>
<tr>
<td>( \omega )</td>
<td>Mode transition parameter</td>
<td>0.01 ms(^{-1} )</td>
</tr>
<tr>
<td>( P_{Ca} )</td>
<td>L-type ( \mathrm{Ca}^{2+} ) channel permeability to ( \mathrm{Ca}^{2+} )</td>
<td>3.125 \times 10^{-4} \text{ cm s}^{-1}</td>
</tr>
<tr>
<td>( P_{K} )</td>
<td>L-type ( \mathrm{Ca}^{2+} ) channel permeability to ( \mathrm{K}^{+} )</td>
<td>5.79 \times 10^{-2} \text{ cm s}^{-1}</td>
</tr>
<tr>
<td>( E_{\text{dref}} )</td>
<td>( E_{\text{d}} ) level that reduces ( P_{Ca} ) by half</td>
<td>(-0.265 \text{ mV} )</td>
</tr>
</tbody>
</table>

### Table 6. Buffering Parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Definition</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>([\text{LTPN}]_{\text{tot}})</td>
<td>Total troponin low-affinity site concentration</td>
<td>70.0 \times 10^{-3} \text{ mmol/L}</td>
</tr>
<tr>
<td>([\text{HTRPN}]_{\text{tot}})</td>
<td>Total troponin high-affinity site concentration</td>
<td>140.0 \times 10^{-3} \text{ mmol/L}</td>
</tr>
<tr>
<td>( K_{\text{Ca}^{2+}} )</td>
<td>( \mathrm{Ca}^{2+} ) on rate for troponin high-affinity sites</td>
<td>20.0 \text{ mmol/L} \text{ ms}^{-1}</td>
</tr>
<tr>
<td>( K_{\text{Ca}^{2+}} )</td>
<td>( \mathrm{Ca}^{2+} ) off rate for troponin high-affinity sites</td>
<td>66.0 \times 10^{-4} \text{ ms}^{-1}</td>
</tr>
<tr>
<td>( K_{\text{Ca}^{2+}} )</td>
<td>( \mathrm{Ca}^{2+} ) on rate for troponin low-affinity sites</td>
<td>40.0 \text{ mmol/L} \text{ ms}^{-1}</td>
</tr>
<tr>
<td>( K_{\text{Ca}^{2+}} )</td>
<td>( \mathrm{Ca}^{2+} ) off rate for troponin low-affinity sites</td>
<td>0.040 ms^{-1}</td>
</tr>
<tr>
<td>([\text{CMDN}]_{\text{tot}})</td>
<td>Total myoplasmic calmodulin concentration</td>
<td>50.0 \times 10^{-3} \text{ mmol/L}</td>
</tr>
<tr>
<td>([\text{CSQN}]_{\text{tot}})</td>
<td>Total NSR calsequestrin concentration</td>
<td>15.0 \text{ mmol/L}</td>
</tr>
<tr>
<td>( K_{\text{Ca}^{2+}} )</td>
<td>( \mathrm{Ca}^{2+} ) half-saturation constant for calmodulin</td>
<td>2.38 \times 10^{-3} \text{ mmol/L}</td>
</tr>
<tr>
<td>( K_{\text{Ca}^{2+}} )</td>
<td>( \mathrm{Ca}^{2+} ) half-saturation constant for calsequestrin</td>
<td>0.8 \text{ mmol/L}</td>
</tr>
</tbody>
</table>
Acknowledgments

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References


<table>
<thead>
<tr>
<th>Variable</th>
<th>Definition</th>
<th>Initial Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>( t )</td>
<td>Time</td>
<td>0.00 ms</td>
</tr>
<tr>
<td>( V )</td>
<td>Membrane potential</td>
<td>-95.87 mV</td>
</tr>
<tr>
<td>( m )</td>
<td>( h_a ) activation gate</td>
<td>( 2.467 \times 10^{-4} )</td>
</tr>
<tr>
<td>( h )</td>
<td>( h_a ) inactivation gate</td>
<td>0.99869</td>
</tr>
<tr>
<td>( j )</td>
<td>( h_s ) slow inactivation gate</td>
<td>0.99887</td>
</tr>
<tr>
<td>( X_{h_1} )</td>
<td>( h_a ) activation gate</td>
<td>0.6935</td>
</tr>
<tr>
<td>( X_{h_2} )</td>
<td>( h_s ) activation gate</td>
<td>( 1.458 \times 10^{-4} )</td>
</tr>
<tr>
<td>( X_{h_3} )</td>
<td>( h_a ) activation gate</td>
<td>( 3.742 \times 10^{-5} )</td>
</tr>
<tr>
<td>( Y_{h_1} )</td>
<td>( h_s ) inactivation gate</td>
<td>1.00</td>
</tr>
<tr>
<td>([Na]^+)</td>
<td>Intracellular Na(^+) concentration</td>
<td>10.00 mmol/L</td>
</tr>
<tr>
<td>([K]^+)</td>
<td>Intracellular K(^+) concentration</td>
<td>159.48 mmol/L</td>
</tr>
<tr>
<td>([Ca^{2+}]_{NSR})</td>
<td>Myoplasmic Ca(^{2+}) concentration</td>
<td>( 8.464 \times 10^{-5} ) mmol/L</td>
</tr>
<tr>
<td>([Ca^{2+}]_{JSR})</td>
<td>Subspace SR Ca(^{2+}) concentration</td>
<td>( 0.2620 ) mmol/L</td>
</tr>
<tr>
<td>([Ca^{2+}]_{SR})</td>
<td>JSR Ca(^{2+}) concentration</td>
<td>( 1.315 \times 10^{-4} ) mmol/L</td>
</tr>
<tr>
<td>( P_{C_1} )</td>
<td>Fraction of channels in state ( P_{C_1} )</td>
<td>0.4929</td>
</tr>
<tr>
<td>( P_{C_2} )</td>
<td>Fraction of channels in state ( P_{C_2} )</td>
<td>6.027 \times 10^{-4}</td>
</tr>
<tr>
<td>( P_{O_1} )</td>
<td>Fraction of channels in state ( P_{O_1} )</td>
<td>( 2.882 \times 10^{-9} )</td>
</tr>
<tr>
<td>( P_{O_2} )</td>
<td>Fraction of channels in state ( P_{O_2} )</td>
<td>0.5065</td>
</tr>
<tr>
<td>( C_2 )</td>
<td>L-type Ca(^{2+}) channel closed: mode normal</td>
<td>0.99802</td>
</tr>
<tr>
<td>( C_1 )</td>
<td>L-type Ca(^{2+}) channel closed: mode normal</td>
<td>( 1.954 \times 10^{-6} )</td>
</tr>
<tr>
<td>( C_0 )</td>
<td>L-type Ca(^{2+}) channel closed: mode normal</td>
<td>0.00</td>
</tr>
<tr>
<td>( O )</td>
<td>L-type Ca(^{2+}) channel open: mode normal</td>
<td>0.00</td>
</tr>
<tr>
<td>( C_{00} )</td>
<td>L-type Ca(^{2+}) channel closed: mode Ca</td>
<td>( 1.9734 \times 10^{-3} )</td>
</tr>
<tr>
<td>( C_{01} )</td>
<td>L-type Ca(^{2+}) channel closed: mode Ca</td>
<td>0.00</td>
</tr>
<tr>
<td>( C_{02} )</td>
<td>L-type Ca(^{2+}) channel closed: mode Ca</td>
<td>0.00</td>
</tr>
<tr>
<td>( C_{03} )</td>
<td>L-type Ca(^{2+}) channel closed: mode Ca</td>
<td>0.00</td>
</tr>
<tr>
<td>( C_{04} )</td>
<td>L-type Ca(^{2+}) channel closed: mode Ca</td>
<td>0.00</td>
</tr>
<tr>
<td>( O_{0} )</td>
<td>L-type Ca(^{2+}) channel open: mode Ca</td>
<td>0.00</td>
</tr>
<tr>
<td>( y )</td>
<td>( h_s ) inactivation gate</td>
<td>0.7959</td>
</tr>
<tr>
<td>([L\text{TPRN}Ca])</td>
<td>Concentration of Ca(^{2+})-bound low-affinity troponin sites</td>
<td>( 5.5443 \times 10^{-5} ) mmol/L</td>
</tr>
<tr>
<td>([H\text{TPRN}Ca])</td>
<td>Concentration of Ca(^{2+})-bound high-affinity troponin sites</td>
<td>( 136.64 \times 10^{-3} ) mmol/L</td>
</tr>
</tbody>
</table>


17. Deleted in proof.


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Raimond L. Winslow, Jeremy Rice, Saleet Jafri, Eduardo Marbán and Brian O'Rourke

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