

Text S1: Model implementation and simulation protocols

## **Impact of sarcoplasmic reticulum calcium release on calcium dynamics and action potential morphology in human atrial myocytes: a computational study**

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## MODEL EQUATIONS

The presented mathematical model is a set of ordinary differential equations that were implemented to the Matlab™ (The MathWorks) environment of technical programming. Simulation results were obtained by numerically integrating the model equations with a stiff ordinary differential equation solver method (ode15s).

Except for the novel  $\text{Ca}^{2+}$  dynamics as well as the new features and modifications described in the *Methods* section, the model equations are as in Nygren *et al.* [1] (see Tables 5-18 in the original ref.). Initial values for the differential variables are listed in Table S1.

Fluxes are in 1 nl volume. Note that cytosol compartments are 1, 2, 3, 4 and ss (subspace) and SR compartments are 1, 2, 3 and 4. The  $\text{Ca}^{2+}$  fluxes between SR and cytosol are as numbered in compartments 1-3, but  $\text{Ca}^{2+}$  flux from SR 4 goes to subspace (see Figure 1).

**$\text{Ca}^{2+}$  fluxes between SR and cytosol in compartments  $x = 1, 2, 3$  and ss (1, 2, 3 and 4 for SR).**

*Serca*

$$k_1 = 10^6 k_4 \quad (1)$$

$$k_2 = k_1 K_{mf}^2 \quad (2)$$

$$k_3 = \frac{k_4}{K_{mr}^2} \quad (3)$$

$$J_{\text{SERCASRx}} = \left\{ -k_3 [\text{Ca}^{2+}]_{\text{SRx}}^2 (c_{\text{pumps}} - [\text{SERCACa}^{2+}]_x) + k_4 [\text{SERCACa}^{2+}]_x \right\} V_x \quad (4)$$

$$J_{x\text{SERCA}} = \left\{ k_1 [\text{Ca}^{2+}]_{\text{ix}}^2 (c_{\text{pumps}} - [\text{SERCACa}^{2+}]_x) - k_2 [\text{SERCACa}^{2+}]_x \right\} V_x \quad (5)$$

$$\frac{d[\text{SERCACa}^{2+}]_x}{dt} = \frac{J_{x\text{SERCA}} - J_{\text{SERCASRx}}}{V_x} \quad (6)$$

*RyR*

$$RyR_{\text{SRCax}} = 1 - \frac{1}{1 + e^{\frac{[\text{Ca}^{2+}]_{\text{SRx}} - 0.3}{0.1}}} \quad (7)$$

$$RyR_{a\infty x} = 0.505 - \frac{0.427}{1 + e^{\frac{1000[Ca^{2+}]_{ix} - 0.29}{0.082}}} \quad (8)$$

$$RyR_{o\infty x} = 1 - \frac{1}{1 + e^{\frac{1000[Ca^{2+}]_{ix} - (RyR_{ax} + 0.22)}{0.03}}} \quad (9)$$

$$RyR_{c\infty x} = \frac{1}{1 + e^{\frac{1000[Ca^{2+}]_{ix} - (RyR_{ax} + 0.02)}{0.01}}} \quad (10)$$

$$J_{relx} = v_x RyR_{ox} RyR_{cx} RyR_{SRcax} ([Ca^{2+}]_{SRx} - [Ca^{2+}]_{ix}) v_x \quad (11)$$

$$\frac{dRyR_{ax}}{dt} = \frac{RyR_{a\infty x} - RyR_{ax}}{\tau_{RyRadapt}} \quad (12)$$

$$\frac{dRyR_{ox}}{dt} = \frac{RyR_{o\infty x} - RyR_{ox}}{\tau_{RyRact}} \quad (13)$$

$$\frac{dRyR_{cx}}{dt} = \frac{RyR_{c\infty x} - RyR_{cx}}{\tau_{RyRinact}} \quad (14)$$

*SR  $Ca^{2+}$  leak*

$$J_{SRleakx} = k_{SRleak} ([Ca^{2+}]_{SRx} - [Ca^{2+}]_{ix}) v_x \quad (15)$$

### ***Intracellular and SR $Ca^{2+}$ dynamics***

*Area accessible for diffusion between junctional and non-junctional cytosol*

$$A_{j-nj} = 2\pi r_{junct} l_{cell} \times 0.5 \quad (16)$$

*Distance between centers of junctional and bulk<sub>4</sub> compartment*

$$x_{j-nj} = \frac{0.2}{2} + \frac{\Delta r}{2} \quad (17)$$

*$Ca^{2+}$  diffusion flux between junctional and non-junctional (bulk<sub>4</sub> compartment) cytosol*

$$J_{j-nj} = D_{Ca} \frac{A_{j-nj}}{x_{j-nj}} ([Ca^{2+}]_{ss} - [Ca^{2+}]_{i4}) \quad (18)$$

*Intracellular  $Ca^{2+}$  fluxes*

$$J_{Ca1} = -J_{1SERCA1} + J_{SRleak1} + J_{rel1} \quad (19)$$

$$J_{Ca2} = -J_{1SERCA2} + J_{SRleak2} + J_{rel2} \quad (20)$$

$$J_{Ca3} = -J_{1SERCA3} + J_{SRleak3} + J_{rel3} \quad (21)$$

$$J_{Ca4} = J_{j-nj} \quad (22)$$

$$J_{Cass} = -J_{j-nj} + J_{SRleakss} - J_{ssSERCA} + J_{relss} \quad (23)$$

*SR Ca<sup>2+</sup> fluxes*

$$J_{SRCa1} = J_{SERCASR1} - J_{SRleak1} - J_{rel1} \quad (24)$$

$$J_{SRCa2} = J_{SERCASR2} - J_{SRleak2} - J_{rel2} \quad (25)$$

$$J_{SRCa3} = J_{SERCASR3} - J_{SRleak3} - J_{rel3} \quad (26)$$

$$J_{SRCa4} = J_{SERCASRss} - J_{SRleakss} - J_{relss} \quad (27)$$

*Ca<sup>2+</sup> buffers, x = 1,2,3 and 4*

$$\beta_{ss} = \left[ 1 + \frac{[SLow]K_{dSLlow}}{([Ca^{2+}]_{ss} + K_{dSLlow})^2} + \frac{[SLhigh]K_{dSLhigh}}{([Ca^{2+}]_{ss} + K_{dSLhigh})^2} + \frac{[BCa]K_{dBCa}}{([Ca^{2+}]_{ss} + K_{dBCa})^2} \right]^{-1} \quad (28)$$

$$\beta_{ix} = \left[ 1 + \frac{[BCa]K_{dBCa}}{([Ca^{2+}]_{ix} + K_{dBCa})^2} \right]^{-1} \quad (29)$$

$$\gamma_{ix} = \frac{[BCa]K_{dBCa}}{[Ca^{2+}]_{ix} + K_{dBCa}} \quad (30)$$

$$\beta_{SRx} = \left[ 1 + \frac{[CSQN]K_{dCSQN}}{([Ca^{2+}]_{SRx} + K_{dCSQN})^2} \right]^{-1} \quad (31)$$

*Intracellular Ca<sup>2+</sup> concentration*

$$\frac{d[Ca^{2+}]_{ss}}{dt} = \beta_{ss} \left( \frac{J_{Cass}}{V_{ss}} + \frac{-I_{CaL} - I_{CaB} - I_{CaP} + 2I_{NaCa}}{2V_{ss}F} \right) \quad (32)$$

For the simulation of Ca<sup>2+</sup> in bulk cytosol the general equation

$$\begin{aligned} \frac{\partial [\text{Ca}^{2+}]_i}{\partial t} = & \beta_i (D_{Ca} + \gamma_i D_{CaBm}) \nabla^2 [\text{Ca}^{2+}]_i \\ & - \frac{2\beta_i \gamma_i D_{CaBm}}{K_{dBCa} + [\text{Ca}^{2+}]_i} \nabla [\text{Ca}^{2+}]_i \cdot \nabla [\text{Ca}^{2+}]_i + J_{Ca} \beta_i \end{aligned} \quad (33)$$

is approximated with radial symmetry to following ODEs:

$$\mathbf{j} = [1 \ 2 \ 3 \ 4] \quad (34)$$

for  $x = 1$

$$\begin{aligned} \frac{d[\text{Ca}^{2+}]_{ix}}{dt} = & \beta_{ix} (D_{Ca} + \gamma_{ix} D_{CaBm}) \left( \frac{[\text{Ca}^{2+}]_{ix+1} - [\text{Ca}^{2+}]_{ix}}{\Delta r^2} \right. \\ & \left. + \frac{[\text{Ca}^{2+}]_{ix+1} - [\text{Ca}^{2+}]_{ix}}{2j_x \Delta r^2} \right) - \frac{2\beta_{ix} \gamma_{ix} D_{CaBm}}{K_{dBCa} + [\text{Ca}^{2+}]_{ix}} \\ & \times \left( \frac{[\text{Ca}^{2+}]_{ix+1} - [\text{Ca}^{2+}]_{ix}}{2\Delta r} \right)^2 + \frac{J_{Cax}}{V_x} \beta_{ix} \end{aligned} \quad (35)$$

for  $x = 2, 3$

$$\begin{aligned} \frac{d[\text{Ca}^{2+}]_{ix}}{dt} = & \beta_{ix} (D_{Ca} + \gamma_{ix} D_{CaBm}) \left( \frac{[\text{Ca}^{2+}]_{ix+1} - 2[\text{Ca}^{2+}]_{ix} + [\text{Ca}^{2+}]_{ix-1}}{\Delta r^2} \right. \\ & \left. + \frac{[\text{Ca}^{2+}]_{ix+1} - [\text{Ca}^{2+}]_{ix-1}}{2j_x \Delta r^2} \right) - \frac{2\beta_{ix} \gamma_{ix} D_{CaBm}}{K_{dBCa} + [\text{Ca}^{2+}]_{ix}} \\ & \times \left( \frac{[\text{Ca}^{2+}]_{ix+1} - [\text{Ca}^{2+}]_{ix-1}}{2\Delta r} \right)^2 + \frac{J_{Cax}}{V_x} \beta_{ix} \end{aligned} \quad (36)$$

for  $x = 4$

$$\begin{aligned} \frac{d[\text{Ca}^{2+}]_{ix}}{dt} = & \beta_{ix} (D_{Ca} + \gamma_{ix} D_{CaBm}) \left( \frac{-[\text{Ca}^{2+}]_{ix} + [\text{Ca}^{2+}]_{ix-1}}{\Delta r^2} \right. \\ & \left. + \frac{[\text{Ca}^{2+}]_{ix} - [\text{Ca}^{2+}]_{ix-1}}{2j_x \Delta r^2} \right) - \frac{2\beta_{ix} \gamma_{ix} D_{CaBm}}{K_{dBCa} + [\text{Ca}^{2+}]_{ix}} \\ & \times \left( \frac{[\text{Ca}^{2+}]_{ix} - [\text{Ca}^{2+}]_{ix-1}}{2\Delta r} \right)^2 + \frac{J_{Cax}}{V_x} \beta_{ix} \end{aligned} \quad (37)$$

Similarly, for the simulation of  $\text{Ca}^{2+}$  in SR the general equation

$$\frac{\partial [\text{Ca}^{2+}]_{\text{SR}}}{\partial t} = \beta_{\text{SR}} D_{Ca\text{SR}} \nabla^2 [\text{Ca}^{2+}]_{\text{SR}} + J_{\text{SRCa}} \beta_{\text{SR}} \quad (38)$$

is approximated with radial symmetry to following ODEs:

for  $x = 1$

$$\begin{aligned} \frac{d[Ca^{2+}]_{SRx}}{dt} = & \beta_{SRx} D_{CaSR} \left( \frac{[Ca^{2+}]_{SRx+1} - [Ca^{2+}]_{SRx}}{\Delta r^2} + \frac{[Ca^{2+}]_{SRx+1} - [Ca^{2+}]_{SRx}}{2j_x \Delta r^2} \right) \\ & + \frac{J_{SRCax}}{V_{SRx}} \beta_{SRx} \end{aligned} \quad (39)$$

for  $x = 2, 3$

$$\begin{aligned} \frac{d[Ca^{2+}]_{SRx}}{dt} = & \beta_{SRx} D_{CaSR} \left( \frac{[Ca^{2+}]_{SRx+1} - 2[Ca^{2+}]_{SRx} + [Ca^{2+}]_{SRx-1}}{\Delta r^2} \right. \\ & \left. + \frac{[Ca^{2+}]_{SRx+1} - [Ca^{2+}]_{SRx-1}}{2j_x \Delta r^2} \right) + \frac{J_{SRCax}}{V_{SRx}} \beta_{SRx} \end{aligned} \quad (40)$$

for  $x = 4$

$$\begin{aligned} \frac{d[Ca^{2+}]_{SRx}}{dt} = & \beta_{SRx} D_{CaSR} \left( \frac{-[Ca^{2+}]_{SRx} + [Ca^{2+}]_{SRx-1}}{\Delta r^2} \right. \\ & \left. + \frac{[Ca^{2+}]_{SRx} - [Ca^{2+}]_{SRx-1}}{2j_x \Delta r^2} \right) + \frac{J_{SRCax}}{V_{SRx}} \beta_{SRx} \end{aligned} \quad (41)$$

*Accessible volumes of cytosol and SR compartments*

$$V_x = \pi l_{cell} [(j_x \Delta r)^2 - (j_{x-1} \Delta r)^2] \times 10^{-6} \times 0.5 \quad (42)$$

$$V_{SRx} = 0.0225 V_x \quad (43)$$

## Sarcolemmal ion currents

The SL ion currents were mostly formulated as in the previously published model of human atrial AP [1]. All the major modifications and novel features of the model are described below. Minor adjustments of parameter values are listed in Table S1.  $I_{to}$ ,  $I_{sus}$ ,  $I_{KS}$ ,  $I_{Kr}$ ,  $I_{Nab}$  and  $I_{NaK}$  were implemented as in [1].

### $Ca^{2+}$ current

The  $I_{CaL}$  was formulated as

$$I_{CaL} = g_{CaL} df_{ca} f_1 f_2 (V - E_{Ca,app}) \quad (44)$$

The time-constant for gate  $f_1$  was fitted to the same data as previously [1,2]. However, as shown in Figure S1A, it was adjusted to have larger values in the membrane voltage range

of -40 and -10 mV; an approach that has been used in other models as well [3]. This modification of the  $I_{CaL}$  formulation was necessary to reduce the overly large tendency of the model to produce early after-depolarizations due to re-activation of  $I_{CaL}$ . The steady-state curve for the  $Ca^{2+}$ -dependent inactivation gate was formulated as

$$f_{ca,\infty} = 1 - \frac{1}{1 + \left( \frac{0.001}{[Ca^{2+}]_{i,ss}} \right)^2} \quad (45)$$

The time-constant for  $f_{ca,\infty}$  was adopted from [4]; see Figure S1B, upper panel. As shown in the lower panel of Figure S1B, this formulation reproduces qualitatively the result that blocking intracellular  $Ca^{2+}$  release decreases the rate of L-type calcium channel inactivation significantly in human atrial myocytes [5]. The channel conductance,  $g_{CaL}$ , was adjusted to 25.3125 nS to reproduce a -12 pA/pF peak current [6,7,8] (Figure S1C) and proper  $Ca^{2+}$  intrusion via L-type  $Ca^{2+}$  channels [5]. See Results for further details.

#### *Hyperpolarization-activated inward $K^+$ current*

The hyperpolarization-activated inward current ( $I_h$ ) was not included in the SL currents of the original Nygren model. Recent experiments indicate that it could have a significant role in atrial myocytes [9,10,11,12], so it was added according to a previously published formulation [12]. The curves for steady-state activation and time-constant were fitted to experimental data [12], as shown in Figure S2. The current equations were implemented according to the formulation of Zorn-Pauly et al. [12] (see Chapter 2.3 for equations in the original ref).

## **MODEL VARIANTS**

To compare the role of SR  $Ca^{2+}$  dynamics in the modulation of action potential duration (APD) to other recently suggested mechanisms, we implemented two additional variants of the myocyte model that included:

- (1) a subsarcolemmal  $Na^+$  compartment and
- (2) an updated scheme of  $K^+$  currents ( $I_{to}$  and  $I_{sus}$ ).

The description of the subsarcolemmal ‘fuzzy’ space for  $Na^+$  corresponds to the recent *in vitro* findings from ventricular myocytes [13,14,15,16]. The  $Na^+$  diffusion coefficient and buffering capacity were set according to the estimates of Despa et al. [15] and Grandi et al. [17], respectively. The size of the subsarcolemmal ‘fuzzy’ space is the same as the corresponding  $Ca^{2+}$  compartment. The area accessible for diffusion between junctional and cytosolic compartments, the distance between centers of junctional and cytosolic

compartment, and the  $\text{Na}^+$  diffusion flux between the junctional and cytosolic compartment are calculated as for  $\text{Ca}^{2+}$  in equations (16–18).

The  $\text{K}^+$  ion currents ( $I_{\text{to}}$  and  $I_{\text{sus}}$ ) were updated according to a recently published human atrial model of Maleckar et al. [18]. The implemented is based on the description in the appendix of the original publication.

In both model variants, #1 and #2, the maximum conductance of inwardly rectifying  $\text{K}^+$  current ( $g_{\text{K1}}$ ) was decreased by 4% and 10%, respectively, to adjust  $\text{APD}_{90}$  at 1Hz pacing to a comparable level.

## **Simulation protocols**

As suggested originally by Hund et al. [19], we implemented a conservative stimulation scheme, in which the stimulus current is carried by  $\text{K}^+$  ions. In pacing experiments, the normal stimulus was a rectangular current pulse that was repeated according to the pacing frequency.

In quiescent experiments, one stimulus pulse was applied at the beginning of the simulation and the myocyte was then left undisturbed for a time period that was required for the model to reach a steady-state.



## REFERENCES

1. Nygren A, Fiset C, Firek L, Clark JW, Lindblad DS, et al. (1998) Mathematical model of an adult human atrial cell - The role of  $K^+$  currents in repolarization. *Circ Res* 82: 63-81.
2. Li GR, Nattel S (1997) Properties of human atrial  $I_{Ca}$  at physiological temperatures and relevance to action potential. *Am J Physiol Heart Circ Physiol* 41: H227-H235.
3. ten Tusscher KHWJ, Panfilov AV (2006) Alternans and spiral breakup in a human ventricular tissue model. *American Journal of Physiology-Heart and Circulatory Physiology* 291: H1088-H1100.
4. ten Tusscher KHWJ, Noble D, Noble PJ, Panfilov AV (2004) A model for human ventricular tissue. *American Journal of Physiology-Heart and Circulatory Physiology* 286: H1573-H1589.
5. Hatem SN, Benardeau A, RuckerMartin C, Marty I, deChamisso P, et al. (1997) Different compartments of sarcoplasmic reticulum participate in the excitation-contraction coupling process in human atrial myocytes. *Circulation Research* 80: 345-353.
6. Nygren A, Fiset C, Firek L, Clark JW, Lindblad DS, et al. (1998) Mathematical model of an adult human atrial cell - The role of  $K^+$  currents in repolarization. *Circulation Research* 82: 63-81.
7. Li GR, Nattel S (1997) Properties of human atrial  $I_{Ca}$  at physiological temperatures and relevance to action potential. *American Journal of Physiology-Heart and Circulatory Physiology* 41: H227-H235.
8. Ouadid H, Albat B, Nargeot J (1995) Calcium Currents in Diseased Human Cardiac-Cells. *Journal of Cardiovascular Pharmacology* 25: 282-291.
9. Hoppe UC, Beuckelmann DJ (1998) Characterization of the hyperpolarization-activated inward current in isolated human atrial myocytes. *Cardiovasc Res* 38: 788-801.
10. Porciatti F, Pelzmann B, Cerbai E, Schaffer P, Pino R, et al. (1997) The pacemaker current  $I_f$  in single human atrial myocytes and the effect of  $\beta$ -adrenoceptor and  $A_1$ -adenosine receptor stimulation. *Br J Pharmacol* 122: 963-969.
11. El Chemaly A, Magaud C, Patri S, Jayle C, Guinamard R, et al. (2007) The Heart Rate-Lowering Agent Ivabradine Inhibits the Pacemaker Current  $I_f$  in Human Atrial Myocytes. *J Cardiovasc Electrophysiol* 18: 1190-1196.
12. Zorn-Pauly K, Schaffer P, Pelzmann B, Lang P, Machler H, et al. (2004)  $I_f$  in left human atrium: a potential contributor to atrial ectopy. *Cardiovasc Res* 64: 250-259.
13. Wendt-Gallitelli MF, Voigt T, Isenberg G (1993) Microheterogeneity of Subsarcolemmal Sodium Gradients. *Electron Probe Microanalysis in Guinea-Pig Ventricular Myocytes*. *J Physiol* 472: 33-44.
14. Fujioka Y, Matsuoka S, Ban T, Noma A (1998) Interaction of the  $Na^+-K^+$  pump and  $Na^+-Ca^{2+}$  exchange via  $[Na^+]_i$  in a restricted space of guinea-pig ventricular cells. *J Physiol* 509: 457-470.
15. Despa S, Bers DM (2003)  $Na/K$  pump current and  $[Na]_i$  in rabbit ventricular myocytes: Local  $[Na]_i$  depletion and  $Na$  buffering. *Biophys J* 84: 4157-4166.
16. Verdonck F, Mubagwa K, Sipido KR (2004)  $[Na^+]_i$  in the subsarcolemmal 'fuzzy' space and modulation of  $[Ca^{2+}]_i$  and contraction in cardiac myocytes. *Cell Calcium* 35: 603-612.
17. Grandi E, Pasqualini FS, Puglisi JL, Bers DM (2009) A Novel Computational Model of the Human Ventricular Action Potential and  $Ca$  transient. *Biophys J* 96: 664a-665a.
18. Maleckar MM, Greenstein JL, Giles WR, Trayanova NA (2009)  $K^+$  current changes account for the rate dependence of the action potential in the human atrial myocyte. *Am J Physiol Heart Circ Physiol* 297: H1398-1410.

19. Hund TJ, Kucera JP, Otani NF, Rudy Y (2001) Ionic charge conservation and long-term steady state in the Luo-Rudy dynamic cell model. *Biophys J* 81: 3324-3331.