Supplement for

A model of cellular cardiac-neural coupling capturing the sympathetic control of sinoatrial node excitability in normotensive and hypertensive rats

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Supplement

The model equations and parameter values for simulating the sympathetic control of the cardiac pacemaker in both of WKY and SHR in this study are listed below. These include the pacemaker activity with specific ion channels function and Ca handling elements, dynamic β-adrenergic modulation on the excitation of SA node, ionic activity of sympathetic varicosity with NE release dynamic and the coupling between varicosity model and SA node model.

Abbreviations

AP	action potential
APA	action potential amplitude
APD_{50}	action potential duration at 50% of the amplitude
cAMP	cyclic adenosine monophosphate
cGMP	cyclic guanosine monophosphate
CL	circle length
ER	endoplasmic reticulum
HR	heart rate
MDP	maximum diastolic potential
MOP	maximum overshoot potential
NE	norepinephrine
NET	NE transporter
PDE2	phosphodiesterase 2
PKA	protein kinase A
RyRs	ryanodine receptors
SERCA	sarcoplasmic reticulum Ca^{2+}-ATPase
SHR	spontaneous hypertensive rat
SNS	sympathetic nervous stimulation
Vmax	maximum upstroke velocity
WKY	wistar Kyoto rat

SA node model development

The eletrophysiological framework employed in our model were modified from the previous rabbit SA node models (1,2) and parameterised on the basis of measurements from rat SA node preparations recorded at the temperature of 35-37 by Satoh, 2003a,b; Shinagawa et al., 2000 (3-5).

Geometry considerations

For the adult rat SA node model, the cell volume was set to be 3.2 pL, which falls in the range of rat SA node according to the study of Silvana et al., 2002 (6).

There are no data of rat SA node available for the volumes of cytoplasmic space, subspace, junctional sarcoplasmic reticulum (SR) and network SR. Since the cell size we used in the model is very close to that of rabbit SA node, we used the same values as Kurata's model (2) for these parameters, which were adopted from the model of Demir et al., 1994(7) based on the data of ventricular myocytes.

The recorded cell capacitance of rat SA node is reported across a wide range from 28 to 158 pF as a result of obtaining measurements from different regions from the SA node (5, 8). In line with our goal of developing a model of primary pacemaking cell located in the central region of SA node, we chose the smaller value of 32 pF.

Equations for membrane action potential and individual ion currents

Membrane potential (V) is mathematically described using the standard equation of charge-conservation

$$\frac{dV}{dt} = \frac{-(I_{CaL} + I_{CaT} + I_{Kr} + I_{Ks} + I_{st} + I_{to} + I_{sus} + I_f + I_{K,Ach} + I_{B,Na} + I_{NaK} + I_{NaCa})}{C_m}, \quad (1)$$

where C_m is the cell capacitance. I_{CaL} and I_{CaT}, the inward L type and T type Ca^{2+} currents; I_{Kr} and I_{Ks}, the rapid and slow delayed rectifier K^+ currents; I_{to}, Ca^{2+}-independent transient outward K^+ current; I_{st}, the sustained inward current (carried by Na^+); I_{sus}, the steady-state outward K^+ current; I_f is the hyperpolarization-activated

current; $I_{K,Ach}$, the muscarinic K^+ channel current; $I_{b,Na}$, the background inward Na^+ current; I_{NaK}, the Na^+-K^+ pump current; I_{NaCa}, the Na^+-Ca^{2+} exchanger current.

I_{Na}

The fast inward Na^+ current initiates the upstroke of action potential in ventricle cell, but is commonly thought to be absent in primary SA node cells. The current density of I_{Na} in the central SA node region is negligibly small compared with the peripheral SA node region (9). In rat SA node it was confirmed that the rapid sodium current was weakly involved in the spontaneous beating (10), so no I_{Na} is included in the model.

I_{CaL}

The equations for L type Ca^{2+} current is modified from Rabbit SA node model developed by Kuruta (2) with parameters of the half activation voltage and the slope factor adjusted to fit the voltage clamp data of rat SA node cells, which were isolated from the right atria of Wistar rat. In the experiments, I_{CaL} was obtained by voltage clamp steps of 300 ms duration from a -40 mV holding potential to test potential between -40 and 50 mV at 35-37 ^oC. Using the same protocol of voltage clamping, the normalized I-V relation produced by our model is plotted in the Figure 2A and the simulated current transients are plotted in the Figure 3A compared to the experimental data (superimposed small print, (8)). In addition, the value of g_{CaL} (maximum current conductance) was increased to 0.7 ns/pF to yield a peak current value of 13 pA at 0 mV consistent with experimental observations (5, 8).

I_{Kr}

The rapid delayed rectifier K^+ current was formulated on the basis of the equations used in Zhang's rabbit SA node model (1). Satoh (11) measured the difference between the peak current and the zero current as the amplitude of the I_{Kr} tail current of rat SA node cell at 36 ^oC, the normalized I-V curve obtained by a set of testing potentials of 200 ms duration from -40 to 50 mV was plotted in Figure 2B and the current transients are plotted in Figure 3B. To fit rat SA node experimental data the half activation and inactivation voltage of I_{Kr} and the slope factors were modified. The Figure 2B also shows the normalized simulated I-V curve under the same voltage clamp protocol is in good agreement with the experimental data (11).

I_{Ks}

The formulations for the slow delayed rectifier K^+ current were derived from Zhang's SA node model (1), and parameterised through the parameters of the opening rate and closing rate constants. The modification was based on the measurements of I_{Ks} tail current in rat SA node cell (11). The experimental and computational curve of the normalized relationship between the peak I_{Ks} tail currents and the testing potentials of 1 s duration from -40 to 50 mV are plotted in the Figure 2C, demonstrating a close agreement. The model generated I_{Ks} current transients are shown in the Figure 3C.

I_{st}

I_{st}, the sustained inward current (carried by Na^+), is the key current in the generation of diastolic depolarization (12, 13). The equations for I_{st} are derived from Shinagawa's study of rat SA node cell with the capacitance of 32 pF. The model parameters for I_{st} are modified with E_{st} was set to 18 mV and g_{st} set to 0.03 ns/pF to yield a similar peak current in our model as experimental observation (5).

I_f

The hyperpolarization-activated current is assumed to be carried by Na^+ and K^+. The formulation of I_f was based on Zhang's SA node model (1), then again fitted to the experimental data of Shinagawa et al., 2000 (5). In the experiments of rat SA node cells, I_f was induced by various testing pulses of 300 ms from -160 to -60 mV at 35-37 ^oC, its activation threshold is about -90 mV in rat SA node which is much more negative than -60—-70 mV in rabbit SA node (14). Such a negative activation potential means that I_f may not contribute to the spontaneous depolarization under normal condition in rat SA node cell. The normalized I-V curves from experimental recordings (5) and simulations were plotted in Figure 2D, they are showing a good agreement. The simulated current transients are plotted in the Figure 3D compared to the experimental data (superimposed small print (4)).

$I_{b,Na}$

Na^+-dependent background current is a small inward current, which contributes a gradual membrane depolarization during the pacemaker potential (7, 15). $I_{b,Na}$ is hypothesised play a potential role during the variation of the other pacemaking

currents in SA node cell (16).Due to the shortage of experimental data for rat SA node, the equations of the rabbit SA node model (2) were used unchanged.

Other currents and Ca^{2+} handling

I_{CaT}, I_{to}, I_{sus}, $I_{K,Ach}$, I_{NaK}, I_{NaCa} and cytosolic Ca^{2+} make variable contributions to the action potential, but there is no evidence to show that they play substantial roles in the process of spontaneous depolarization (3, 17-21). A sensitivity analysis in the model shows that none of those currents significantly effects AP morphology and the blocking of these individual current does not stop spontaneous activation. These sensitivity results are shown in the supplement as a table due to the page limitation.

Furthermore, as there is no experimental rat data available for these components and so the models for each component are reproduced from Zhang's and Kuruta's rabbit SA node models (1, 2).

Model parameters

SA node

$[Ca^{2+}]_o$ (mM)	2
C_m (pF)	32
$[CM]_{tot}$ (mM)	0.045
$[CQ]_{tot}$ (mM)	10
d_{NaCa}	0.0001
$E_{Ca,L}$ (mV)	45
$E_{Ca,T}$ (mV)	45
E_k (mV)	(RT/F)*ln([K]o/[K]i)
E_{ks} (mV)	(RT/F)*ln([K]o+0.12*[Na]o)/([Na]i+0.12*[K]i)
E_{Na} (mV)	(RT/F)*ln([Na]o/[Na]i)
E_{st} (mV)	18
F ($C\cdot M^{-1}$)	96,487
g_{bNa} (nS/pF)	0.003
g_{CaL} (nS/pF)	0.7
g_{CaT} (nS/pF)	0.458

g_f	(nS/pF)	0.357
g_{Kr}	(nS/pF)	$0.025*([K]o)^{0.59}$
g_{Ks}	(nS/pF)	0.0259
g_{st}	(nS/pF)	0.03
g_{sus}	(nS/pF)	0.01
g_{to}	(nS/pF)	0.18
$I_{NaK\text{-}max}$	(pA/pF)	3.6
k_{bCM}	(ms^{-1})	0.542
k_{bCQ}	(ms^{-1})	0.445
k_{bTC}	(ms^{-1})	0.446
k_{bTMC}	(ms^{-1})	0.00751
k_{bTMM}	(ms^{-1})	0.751
k_{fCM}	($mM^{-1}\cdot ms^{-1}$)	227.7
k_{fCQ}	($mM^{-1}\cdot ms^{-1}$)	0.534
k_{fTC}	($mM^{-1}\cdot ms^{-1}$)	88.8
k_{fTMC}	($mM^{-1}\cdot ms^{-1}$)	227.7
k_{fTMM}	($mM^{-1}\cdot ms^{-1}$)	2.277
$K_{m,fCa}$	(mM)	0.00035
$K_{m,Kp}$	(mM)	1.4
$K_{m,Nap}$	(mM)	14
k_{NaCa}	(mM^{-4})	0.0005
K_{rel}	(mM)	0.0012
K_{up}	(mM)	0.0006
$[K^+]_o$	(mM)	5.4
$[Mg^+]_i$	(mM)	2.5
$[Na^+]_o$	(mM)	140
P_{rel}	(ms^{-1})	5
P_{up}	(mM/ms)	0.005
R	($J\cdot mol^{-1}\cdot K^{-1}$)	8.314472
T	(K)	310
$[TC]_{tot}$	(mM)	0.031
$[TMC]_{tot}$	(mM)	0.062

V_{cell} (pl)	3.5
V_i (pl)	$0.46V_{cell}$-Vsub
V_{rel} (pl)	$0.0012V_{cell}$
V_{sub} (pl)	$0.01V_{cell}$
V_{up} (pl)	$0.0116V_{cell}$
τ_{tr} (ms)	60
τ_{diff_Ca} (ms)	0.04
y_{NaCa}	0.5
alpha_fCa (ms^{-1})	0.035
beta_fCa ($mM^{-1}*ms^{-1}$)	60

Neuron

b	0.25
C_m (pF)	240
E_{Ca} (mV)	145
E_K (mV)	-93
E_{Na} (mV)	40
gama (ms^{-1})	6
g_{Na} (μS)	9.28
P_{Ca} (cm/ms)	1.1×10^{-16}
g_{KV} (μS)	0.44
g_A (μS)	2.11
g_{Kcf} (μS)	0.15
g_{Kcs} (μS)	0.22
K_{on} ($mM^{-1}ms^{-1}$)	90
K_{off} (ms^{-1})	9.5
V_Z (L)	10×10^{-17}

Cleft

P_{up} (ms^{-1})	10000
V_{cleft} (L)	1.5×10^{-15}

SHR MODEL

The values of the parameters P_{Ca} (1.1×10^{-16}cm/ms) and PDE_{tot} (39.8×10^{-6} mM) in the WKY model were changed for the SHR model as below:

P_{Ca} =1.54×10^{-16} cm/ms

PDE_{tot} =29.45×10^{-6} mM

Equations

SA node Sarcolemmal Ionic Current

L-type Ca^{2+} current

$$I_{Ca,L}= f\_PKA\_L \times g_{Ca,L} \times (V - E_{CaL}) \times d \times f \times fCa \quad (1)$$

$$\bar{d} = \frac{1}{1+e^{-(V+12.1+V\_SHIFT\_L)/6}} \quad (2)$$

$$\tau_d = \frac{1}{\alpha_d + \beta_d} \quad (3)$$

$$\alpha_d = \frac{-0.02839\times(V+35)}{e^{-(V+35)/2.5}-1} - \frac{0.0849\times V}{e^{-V/4.808}-1} \quad (4)$$

$$\beta_d = \frac{0.0143\times(V-5)}{e^{(V-5)/2.5}-1} \quad (5)$$

$$\frac{dd}{dt} = \frac{\bar{d}-d}{\tau_d} \quad (6)$$

$$\bar{f} = \frac{1}{1+e^{(V+30+V\_SHIFT\_L)/5}} \quad (7)$$

$$\tau_f = 44.3 + 257.1\times e^{-(\frac{V+32.5}{13.9})^2} \quad (8)$$

$$\frac{df}{dt} = \frac{\bar{f}-f}{\tau_f} \quad (9)$$

$$\alpha_{fCa} = Km_{fCa} \times \beta_{fCa} \quad (10)$$

$$\bar{f}_{Ca} = \frac{Km_{fCa}}{Km_{fCa}+[Ca]_{sub}} \quad (11)$$

$$\tau_{fCa} = \frac{\overline{fCa}}{\alpha_{fCa}} \tag{12}$$

$$\frac{df_{Ca}}{dt} = \frac{\overline{f_{Ca}} - f_{Ca}}{\tau_{fCa}} \tag{13}$$

T-type Ca^{2+} current

$$I_{Ca,T} = g_{Ca,T} \times (V - E_{CaT}) \times d \times f \tag{14}$$

$$\overline{d} = \frac{1}{1 + e^{-(V+26.3)/6}} \tag{15}$$

$$\tau_d = \frac{1}{1.068 \times e^{\frac{V+26.3}{30}} + 1.068 \times e^{\frac{-(V+26.3)}{30}}} \tag{16}$$

$$\frac{dd}{dt} = \frac{\overline{d} - d}{\tau_d} \tag{17}$$

$$\overline{f} = \frac{1}{1 + e^{(V+61.7)/5.6}} \tag{18}$$

$$\tau_f = \frac{1}{0.0153 \times e^{\frac{-(V+61.7)}{83.3}} + 0.015 \times e^{\frac{V+61.7}{15.38}}} \tag{19}$$

$$\frac{df}{dt} = \frac{\overline{f} - f}{\tau_f} \tag{20}$$

Rapid delayed rectifier K^+ current

$$I_{Kr} = g_{Kr} \times (V - E_K) \times (0.6 \times paF + 0.4 \times paS) \times piy \tag{21}$$

$$\overline{pa} = \frac{1}{1 + e^{-(V+24)/5}} \tag{22}$$

$$\tau_{paS} = \frac{1}{0.0042 \times e^{\frac{V-9}{17}} + 0.00015 \times e^{\frac{-(V-9)}{21.6}}} \tag{23}$$

$$\tau_{paF} = \frac{1}{0.0372 \times e^{\frac{V-9}{15.9}} + 0.00096 \times e^{\frac{-(V-9)}{22.5}}} \tag{24}$$

$$\frac{dpaS}{dt} = \frac{\overline{paS} - paS}{\tau_{paS}} \tag{25}$$

$$\frac{dpaF}{dt} = \frac{\overline{paF} - paF}{\tau_{paF}} \quad (26)$$

$$\overline{piy} = \frac{1}{1+e^{(V+9.6)/10.1}} \quad (27)$$

$$\tau_{piy} = 2 \quad (28)$$

$$\frac{dpiy}{dt} = \frac{\overline{piy} - piy}{\tau_{piy}} \quad (29)$$

Slow delayed rectifier K^+ current

$$I_{Ks} = f\_Ks \times g_{Ks} \times (V - E_{Ks}) \times n^2 \quad (30)$$

$$\bar{n} = \frac{\alpha_n}{\alpha_n + \beta_n} \quad (31)$$

$$\tau_n = \frac{1}{\alpha_n + \beta_n} \quad (32)$$

$$\alpha_n = \frac{0.014}{1+e^{-(V-40+SHIFT\_Ks)/45}} \quad (33)$$

$$\beta_n = 0.001 \times e^{\frac{-(V+SHIFT\_Ks)}{22}} \quad (34)$$

$$\frac{dn}{dt} = \frac{\bar{n} - n}{\tau_n} \quad (35)$$

Ca^{2+}-independent transient outward K^+ current

$$I_{to} = g_{to} rq(V - E_K) \quad (36)$$

$$\bar{r} = \frac{1}{1+e^{-(V-19.3)/15}} \quad (37)$$

$$\bar{q} = \frac{1}{1+e^{(V+49)/13}} \quad (38)$$

$$\tau_r = \frac{14.405}{1.037e^{0.09(V+30.61)} + 0.369e^{-0.12(V+23.84)}} + 2.7535 \quad (39)$$

$$\tau_q = \frac{39.102}{0.57 \times e^{-0.08(V+44)} + 0.065 \times e^{0.1(V+45.93)}} + 6.06 \quad (40)$$

$$\tau_{Sslow} = 3.7e^{-(V+70.0/30.0)^2} + 0.035 \quad (41)$$

$$\frac{dr}{dt} = \frac{\bar{r} - r}{\tau_r} \tag{42}$$

$$\frac{dq}{dt} = \frac{\bar{q} - q}{\tau_q} \tag{43}$$

Steady-state outward K^+ current

$$I_{sus} = g_{sus} r (V - E_K) \tag{44}$$

$$\bar{r} = \frac{1}{1 + e^{-(V-19.3)/15}} \tag{45}$$

$$\tau_r = \frac{14.405}{1.037 e^{0.09(V+30.61)} + 0.369 e^{-0.12(V+23.84)}} + 2.7535 \tag{46}$$

$$\frac{dr}{dt} = \frac{\bar{r} - r}{\tau_r} \tag{47}$$

Hyperpolarization-activated current

$$I_f = g_f \times (I_{fNa} + I_{fK}) \tag{48}$$

$$I_{fNa} = g_{fNa} \times (V - E_{Na}) \times y^2 \tag{49}$$

$$I_{fK} = g_K \times (V - E_K) \times y^2 \tag{50}$$

$$\bar{y} = \frac{1}{1 + e^{(V+100-V\_shift)/13.5}} \tag{51}$$

$$\tau_y = \frac{0.7166529}{e^{-(V+425.5)/45.302} + e^{(V-73.08)/19.231}} \tag{52}$$

$$\frac{dy}{dt} = \frac{y_\infty - y}{\tau_y} \tag{53}$$

Sustained inward current

$$I_{st} = f\_PKA\_st \times g_{st} \times (V - E_{st}) \times qa \times qi \tag{54}$$

$$\overline{qa} = \frac{1}{1 + e^{-(V+57)/5}} \tag{55}$$

$$\tau_{qa} = \frac{1}{\alpha_{qa} + \beta_{qa}} \tag{56}$$

$$\alpha_{qa} = \frac{1}{0.15 \times e^{\frac{-V}{11}} + 0.2 \times e^{\frac{-V}{700}}} \tag{57}$$

$$\beta_{qa} = \frac{1}{16 \times e^{\frac{V}{8}} + 15 \times e^{\frac{V}{50}}} \tag{58}$$

$$\frac{dqa}{dt} = \frac{qa_{\infty} - qa}{\tau_{qa}} \tag{59}$$

$$\overline{qi} = \frac{\alpha_{qi}}{\alpha_{qi} + \beta_{qi}} \tag{60}$$

$$\tau_{qi} = \frac{1}{\alpha_{qi} + \beta_{qi}} \tag{61}$$

$$\alpha_{qi} = \frac{1}{3100 \times e^{\frac{V}{13}} + 700 \times e^{\frac{-V}{70}}} \tag{62}$$

$$\beta_{qi} = \frac{1}{95 \times e^{\frac{-V}{10}} + 50 \times e^{\frac{-V}{700}}} + \frac{0.000229}{1 + e^{\frac{-V}{5}}} \tag{63}$$

$$\frac{dqi}{dt} = \frac{qi_{\infty} - qi}{\tau_{qi}} \tag{64}$$

Background sodium current

$$I_{b,Na} = g_{b,Na}(V - E_{Na}) \tag{65}$$

Muscarinic potassium channel current

$$I_{K\_Ach} = g_{K\_Ach} \times (Ki - Ko \times e^{\frac{-VF}{RT}}) \tag{66}$$

Na^+-K^+ pump current

$$I_{NaK} = I_{NaK\_\max} \times (1 + (\frac{Km_{Kp}}{Ko})^{1.2})^{-1} \times (1 + (\frac{Km_{Nap}}{Nai})^{1.3})^{-1} \times (1 + e^{\frac{-(V - E_{Na} + 120)}{30}})^{-1} \tag{67}$$

Na^+-Ca^{2+} exchanger current

$$I_{NaCa} = k_{NaCa} \times \frac{[Na^+]_i^{\,3} \times [Ca^{2+}]_o \times e^{0.03743 V \gamma_{NaCa}} - [Na^+]_o^{\,3} \times [Ca^{2+}]_{sub} \times e^{0.03743 V (\gamma_{NaCa} - 1.0)}}{1.0 + d_{NaCa} \times ([Na^+]_i^{\,3} \times [Ca^{2+}]_o + [Na^+]_o^{\,3} \times [Ca^{2+}]_{sub})} \tag{68}$$

Intracellular Ca^{2+} Dynamics

$$J_{Ca,diff} = \frac{[Ca^{2+}]_{sub} - [Ca^{2+}]_i}{\tau_{diff\_Ca}} \tag{69}$$

$$J_{rel} = P_{rel} \times ([Ca^{2+}]_{rel} - [Ca^{2+}]_{sub}) \times \frac{[Ca^{2+}]_{sub}{}^2}{[Ca^{2+}]_{sub}{}^2 + K_{rel}{}^2} \tag{70}$$

$$J_{up} = P_{up} \times \frac{[Ca^{2+}]_i}{[Ca^{2+}]_i + K_{up}} \tag{71}$$

$$J_{tr} = \frac{[Ca^{2+}]_{up} - [Ca^{2+}]_{rel}}{\tau_{\_tr}} \tag{72}$$

Intracellular ion concentrations

$$\frac{d[Ca^{2+}]_i}{dt} = \frac{J_{Ca\_diff} \times V_{sub} - J_{up} \times V_{up} - I_{CaP}}{V_i} - ([CM]_{tot} \times f_{CMi\_rate} + [TC]_{tot} \times f_{TC\_rate} + [TMC]_{tot} \times f_{TMC\_rate}) \tag{73}$$

$$\frac{d[Ca^{2+}]_{sub}}{dt} = \frac{-(I_{CaL} + I_{CaT} - 2.0I_{NaCa}) \times Cm/(2.0 \times F) + J_{rel} \times V_{rel}}{V_{sub}} - (J_{Ca\_diff} + [CM]_{tot} \times f_{CMs\_rate}) \tag{74}$$

$$\frac{d[Ca^{2+}]_{rel}}{dt} = J_{tr} - J_{rel} - [CQ]_{tot} \times f_{CQ\_rate} \tag{75}$$

$$\frac{d[Ca^{2+}]_{up}}{dt} = J_{up} - J_{tr} \times \frac{V_{rel}}{V_{up}} \tag{76}$$

$$\frac{d[Na^+]_i}{dt} = -(I_{st} + I_{b,Na} + 3I_{NaCa} + 3I_{NaK} + I_{f,Na}) \frac{Cm}{(V_i + V_{sub})F} \tag{77}$$

$$\frac{d[K^+]_i}{dt} = -(I_{Kr} + I_{sus} + I_{K\_Ach} + I_{to} + I_{Ks} + I_{f,K} - 2I_{NaK}) \frac{Cm}{(V_i + V_{sub})F} \tag{78}$$

Ca^{2+} buffering

$$\frac{df_{TC}}{dt} = Kf_{TC} \times [Ca^{2+}]_i \times (1 - f_{TC}) - Kb_{TC} \times f_{TC} \tag{79}$$

$$\frac{df_{TMC}}{dt} = Kf_{TMC} \times [Ca^{2+}]_i \times (1 - f_{TMC} - f_{TMM}) - Kb_{TMC} \times f_{TMC} \tag{80}$$

$$\frac{df_{TMM}}{dt} = Kf_{TMM} \times [Mg^{2+}]_i \times (1 - f_{TMC} - f_{TMM}) - Kb_{TMM} \times f_{TMM} \tag{81}$$

$$\frac{df_{CMi}}{dt} = Kf_{CM} \times [Ca^{2+}]_i \times (1 - f_{CMi}) - Kb_{CM} \times f_{CMi} \quad (82)$$

$$\frac{df_{CMs}}{dt} = Kf_{CM} \times [Ca^{2+}]_{sub} \times (1 - f_{CMs}) - Kb_{CM} \times f_{CMs} \quad (83)$$

$$\frac{df_{CQ}}{dt} = Kf_{CQ} \times [Ca^{2+}]_{rel} \times (1 - f_{CQ}) - Kb_{CQ} \times f_{CQ} \quad (84)$$

Neuron Sarcolemmal Ionic Current

I_{Na_N}

$$I_{Na\_N} = g_{Na} \times m^3 \times h \times (V - E_{Na}) \quad (85)$$

$$\overline{m} = \frac{1}{1 + e^{-(V+36)/7.2}} \quad (86)$$

$$\overline{h} = \frac{1}{1 + e^{(V+53.2)/6.5}} \quad (87)$$

$$\tau_m = \frac{1}{42.98e^{0.08915 \bullet V} + 0.923e^{-0.03351 \bullet V}} + 0.06 \quad (88)$$

$$\tau_h = \frac{(-0.0046 \times V + 0.26) + 50.85}{(1 + e^{-(V+59.46)/7.91}) + (1 + e^{(V+40.94)/1.556})} \quad (89)$$

I_{Ca_N}

$$I_{Ca\_N} = \frac{4F^2VP_{Ca}}{RT} \times \frac{[Ca]i \times e^{(2VF/RT)} - [Ca]o}{e^{(2VF/RT)} - 1} c^2 \times hc \quad (90)$$

$$\overline{c} = \frac{1}{1 + e^{-(V+8.1)/9.85}} \quad (91)$$

$$\tau_C = \frac{1}{0.343 \times e^{(0.0925V)} + 1.88 \times e^{(-0.00732V)}} + 0.1 \quad (92)$$

$$\overline{hc} = \frac{1}{1 + e^{(V+19.91)/4.5}} \quad (93)$$

$$\tau_{hc} = \frac{1}{0.242 \times e^{(0.145V)} + 0.0434 \times e^{(-0.0201V)}} + 17 \quad (94)$$

I_{KV}

$$I_{KV} = g_{KV} \times n \times (V - E_K) \quad (95)$$

$$\overline{n} = \frac{1}{1 + e^{-(V+6.084)/8.007}} \quad (96)$$

$$\tau_n = \frac{1}{0.385 \times e^{(0.101V)} + 8.134 \times 10^{-4} \times e^{(-0.1177V)}} + 2.31 \quad (97)$$

I_A

$$I_A = g_A \times a \times ah \times (V - E_K) \quad (98)$$

$$\overline{a} = \frac{1}{1 + e^{-(V+49.4)/9.9}} \quad (99)$$

$$\tau_a = \frac{1}{3.7 \times e^{(0.03856V)} + 0.05362 \times e^{(-0.04448V)}} + 0.2 \quad (100)$$

$$\overline{ha} = \frac{1}{1 + e^{(V+80.43)/7.07}} \quad (101)$$

$$\tau_{ha} = \frac{(-0.038V + 6.0) + 87.0}{(1 + e^{-(80+V)/5}) + (1 + e^{(55+V)/1.4})} \quad (102)$$

I_{KCa}

$$I_{KV} = g_{kcf} \times kcf \times (V - E_K) + g_{kcs} \times kcs \times (V - E_K) \quad (103)$$

$$\overline{kcf} = \frac{1}{1 + e^{-(V+11.54)/4.99}} \quad (104)$$

$$\tau_{kcf} = \frac{1}{1292 \times e^{(0.274V)} + 0.00359 \times e^{(-0.126V)}} + 1.4 \quad (105)$$

$$\overline{kcs} = \frac{1}{1 + e^{-(V+15.18)/4.0}} \quad (106)$$

$$\tau_{kcs} = \frac{1}{(0.25 \times e^{0.7V}) + (0.00073 \times e^{(-0.144V)})} + (92 - 1.9V) \quad (107)$$

Ca^{2+} binding to vesicles, vesicle fusion and NE release

$$\frac{dV_{Ca}}{dt} = K_{off} \times V_{1Ca} \times b^0 - 5 \times K_{on} \times [Ca^{2+}]_i \times V_{Ca} \quad (108)$$

$$\frac{dV_{1Ca}}{dt} = 5 \times K_{on} \times [Ca^{2+}]i \times V_{ca} + 2K_{off} \times V_{2Ca} \times b^1 - (K_{off} \times b^0 + 4K_{on} \times [Ca^{2+}]_i) \times V_{1Ca} \quad (109)$$

$$\frac{dV_{2Ca}}{dt} = 4 \times K_{on} \times [Ca^{2+}]i \times V_{1ca} + 3K_{off} \times V_{3Ca} \times b^2 - (2K_{off} \times b^1 + 3K_{on} \times [Ca^{2+}]_i) \times V_{2Ca} \quad (110)$$

$$\frac{dV_{3Ca}}{dt} = 3 \times K_{on} \times [Ca^{2+}]i \times V_{2ca} + 4K_{off} \times V_{4Ca} \times b^3 - (3K_{off} \times b^2 + 2K_{on} \times [Ca^{2+}]_i) \times V_{3Ca} \quad (111)$$

$$\frac{dV_{4Ca}}{dt} = 2 \times K_{on} \times [Ca^{2+}]i \times V_{3ca} + 5K_{off} \times V_{2Ca} \times b^4 - (4K_{off} \times b^3 + K_{on} \times [Ca^{2+}]_i) \times V_{4Ca} \quad (112)$$

$$\frac{dV_{5Ca}}{dt} = K_{on} \times V_{4Ca} \times [Ca^{2+}]i - 5 \times K_{off} \times b^4 \times V_{5Ca} \quad (113)$$

$$E = \gamma \times V_{5Ca} \quad (114)$$

β-adrenergic signalling

β-adrenergic receptor module

$$[L_{tot}] = [L] + [LR] + [LRG] \quad (115)$$

$$[G_{tot}] = [G] + [RG] + [LRG] + [G_{\beta\gamma}] \quad (116)$$

$$[R] = [R_{act}] + [LR] + [LRG] - [RG] \quad (117)$$

$$[LR] = \frac{[L] \times [R]}{0.001} \quad (118)$$

$$[LRG] = \frac{[G] \times [LR]}{0.000062} \quad (119)$$

$$[RG] = \frac{[G] \times [R]}{0.033} \quad (120)$$

$$\frac{d[R_{S464}]}{dt} = 0.0000011 \times ([LR] + [LRG]) - 0.0000022 \times [R_{S464}] \quad (121)$$

$$\frac{d[R_{S301}]}{dt} = 0.0036 \times [PKA] \times [R_{act}] - 0.0000002232 \times [R_{S301}] \quad (122)$$

$$[R_{act}] = [R_{tot}] - ([R_{S464}] + [R_{S301}]) \quad (123)$$

$$\frac{dG_\alpha GTP_{tot}}{dt} = 0.016 \times ([RG] + [LRG]) - 0.001 \times [G_\alpha GTP_{tot}] \quad (124)$$

$$\frac{d[G_{\beta\gamma}]}{dt} = 0.016 \times ([RG] + [LRG]) - 1200 \times [G_\alpha GDP] \times [G_{\beta\gamma}] \quad (125)$$

$$\frac{d[G_\alpha GDP]}{dt} = 0.001 \times [G_\alpha GTP_{tot}] - 1200 \times [G_\alpha GDP] \times [G_{\beta\gamma}] \quad (126)$$

cAMP module

$$[AC_{tot}] = [AC] + [G_\alpha GTP\_AC] \tag{127}$$

$$[G_\alpha GTP_{tot}] = [G_\alpha GTP] + [G_\alpha GTP\_AC] \tag{128}$$

$$[G_\alpha GTP\_AC] = [G_\alpha GTP] \times \frac{[AC]}{0.315} \tag{129}$$

$$\frac{d[cAMP_{tot}]}{dt} = \frac{0.0035 \times [AC] \times [ATP_{total}]_i}{1.03 + [ATP_{total}]_i} + \frac{91.0 \times [G_\alpha GTP\_AC] \times [ATP_{total}]_i}{0.315 + [ATP_{total}]_i} - \frac{0.031 \times [PDE] \times [cAMP]}{0.0013 + [cAMP]} \tag{130}$$

PKA activation module

$$[cAMP_{tot}] = [cAMP] + ([ARC] + 2 \times [A_2RC] + 2 \times [A_2R]) \tag{131}$$

$$2 \times [PKA_{tot}] = [RC] + [ARC] + [A_2RC] + [PKA] + [PKA\_PKI] \tag{132}$$

$$[PKA\_PKI] = \frac{[PKI_{tot}] \times [PKA]}{0.001 + [PKA]} \tag{133}$$

$$[A_2R] = [PKA] + [PKA\_PKI] \tag{134}$$

$$[A_2RC] = \frac{[PKA] \times [A_2R]}{0.009} \tag{135}$$

$$[RC] = \frac{0.008 \times [ARC]}{[cAMP]} \tag{136}$$

$$[ARC] = \frac{0.008 \times [A_2RC]}{[cAMP]} \tag{137}$$

B1-adrenergic modulation on SA node cell

I_{CaL}

$$f\_PKA\_L = \frac{17.5 \times ([PKACII] - 0.000008278)}{([PKACII] + 0.0004)} + 1 \tag{138}$$

$$V\_SHIFT\_L = \frac{62.5 \times ([PKACII] - 0.000008278)}{([PKACII] + 0.0004)} \tag{139}$$

I_{st}

$$f\_PKA\_st = \frac{([cAMP] - 0.00002)}{([cAMP] + 0.001 - 0.0002)} + 1 \tag{140}$$

I_f

$$V\_SHIFT\_f = \frac{20 \times ([cAMP] - 0.000224)}{([cAMP] + 0.001)} \quad (141)$$

I_{Ks}

$$f\_Ks = \frac{0.2 \times F\_KCNQ1p}{0.015} + 0.8 \quad (142)$$

$$V\_SHIFT\_Ks = \frac{2.0 \times F\_KCNQ1p}{0.04} - 2.0 \quad (143)$$

J_{up}

$$Kup = \frac{Kup0 \times (1 + 2 \times fracPLB)}{1 + 2 \times fracPLB0} \quad (144)$$

Table1. Simulated effects of the blockade of the membrane currents on the pacemaker activity.

Membrane current blocked	**Change on CL (rat SA node model)**
I_{Cal}	Stop
I_{st}	Stop
I_{Kr}	Stop
I_{Ks}	+6.6%
I_{CaT}	+15.6%
I_f	No effect
$I_{K,ach}$	-3.5%
$I_{b,Na}$	Stop
I_{sus}	+6%
I_{to}	-1.16%

Table 2. Simulated effects of the variations of model components on the AP

Membrane Current	**10% perturbation on the current density**
	CL (%)
I_{CaL}	0.78
I_{st}	2.7
I_{Kr}	0.39
I_{Ks}	0
I_{CaT}	1.17
$I_{K,ach}$	0.78
I_f	0
$I_{b,Na}$	1.13
I_{NaCa}	0.39
I_{NaK}	0.78
I_{sus}	0.39
I_{to}	0

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