An approach to the biological pacemaker engineering based on the bifurcation analysis of a human ventricular cell model

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Abstract—Electronic pacemakers play an important role in the treatment of bradyarrhythmia. Recently, different strategies to initiate pacemaker activity by genetic modification of ionic currents across the cell membrane were developed. These new approaches are called biological pacemaker (BP) engineering. Recent studies on the creation of the BP using mathematical cell models have revealed the mechanism of induction of pacemaker activity and pacemaking current. However, these studies have used the models of a guinea pig ventricular myocyte. In this paper, we use the reduced ten Tusscher-Noble-Noble-Panfilov (TNNP) model of human ventricular myocytes. We analyze the bifurcation structure of the model by varying conductances of ion channels, and examine the method to create a BP. The results show that the human ventricular myocytes can be converted to cardiac pacemaker by suppression of the inward rectifier potassium current.

1. Introduction

A heart plays a physiological role as the pump for the blood circulation by repeating contractions and relaxations regularly. These periodic motions are controlled by electrical signals (action potentials) generated in the sinoatrial node, which is a physiological pacemaker of the heart. Generation of the action potential is related to various ion channels in the cell membrane.

In biological cells, the difference of the ionic concentration between inside and outside of the cells makes the difference of electrical potential (membrane potential). In the cell membrane, there are various ion channels which open and close depending on the membrane potential. Various ions move inwards or outwards the cell membrane through the ionic channels, as a result, action potential is generated.

If the rhythm of sinoatrial node is disrupted by aging or a variety of cardiovascular disorders, the heartbeat can become too slow (bradyarrhythmia). In that case, the implantation of electronic cardiac pacemaker is required. Recently, the new strategies to initiate pacemaker function by genetic modification of ionic currents have developed. These pacemakers are called biological pacemakers (BPs). Physiological experiments have demonstrated that spontaneous action potential is induced by suppressing a specific ionic current (I_{K1}) in the guinea pig ventricular cells [1].

Since the Hodgkin–Huxley equations [2] were published in 1962, various mathematical cell models have been developed [3]. Since it is difficult to analyze the dynamics of membrane potential variations only by physiological experiments, mathematical cell models have been applied to the studies on the BPs. Recent studies on the BPs using cardiac cell models have reveled the mechanism of induction of pacemaker activity [4, 5]. However, these studies have used the models of a guinea pig ventricular myocyte.

This paper uses the reduced version of the ten Tusscher–Noble–Noble–Panfilov (TNNP) model [6] of a human ventricular myocyte, which is described by the nonlinear ordinary differential equations with nine variables. We investigate the bifurcation structure of the reduced TNNP model by varying conductances of ion channels and examine the effect of ionic currents on the pacemaker activity.

2. Reduced ten Tusscher–Noble–Noble–Panfilov Model

The reduced TNNP model is a human ventricular cell model described by the Hodgkin–Huxley-type equations with nine variables. The variation of membrane potential V (mV) is described by

$$\frac{\mathrm{d}V}{\mathrm{d}t} = -\frac{1}{C_\mathrm{m}}(I_\mathrm{stim} + I_\mathrm{Na} + I_\mathrm{CaL} + I_\mathrm{to} + I_\mathrm{Kr} + I_\mathrm{Ks} + I_\mathrm{K1} + I_\mathrm{NaCa} + I_\mathrm{NaK} + I_\mathrm{pCa} + I_\mathrm{pK} + I_\mathrm{bNa} + I_\mathrm{bCa}) \tag{1}$$

where C_m (μF/cm^2) is the membrane capacitance, I_stim is the externally applied stimulus current, I_Na, I_CaL, I_to, I_Kr, I_Ks, I_K1, I_NaCa, I_NaK, I_pCa, I_pK, I_bNa and I_bCa are the ionic currents. These currents are given by the following equations:

$$I_\mathrm{Na} = c_\mathrm{Na} G_\mathrm{Na} m^3 h j (V - E_\mathrm{Na}) \tag{2}$$
$$I_\mathrm{CaL} = c_\mathrm{CaL} G_\mathrm{CaL} d_\infty f_1 f_2 (V - 60) \tag{3}$$
$$I_\mathrm{to} = c_\mathrm{to} G_\mathrm{to} r_\infty s (V - E_\mathrm{K}) \tag{4}$$
$$I_\mathrm{Kr} = c_\mathrm{Kr} G_\mathrm{Kr} \sqrt{\frac{[\mathrm{K}^+]_\mathrm{o}}{5.4}} x_\mathrm{r1} x_\mathrm{r2\infty} (V - E_\mathrm{K}) \tag{5}$$
$$I_\mathrm{Ks} = c_\mathrm{Ks} G_\mathrm{Ks} x_\mathrm{s}^2 (V - E_\mathrm{Ks}) \tag{6}$$
$$I_\mathrm{K1} = c_\mathrm{K1} G_\mathrm{K1} \sqrt{\frac{[\mathrm{K}^+]_\mathrm{o}}{5.4}} x_\mathrm{K1\infty} (V - E_\mathrm{K}) \tag{7}$$

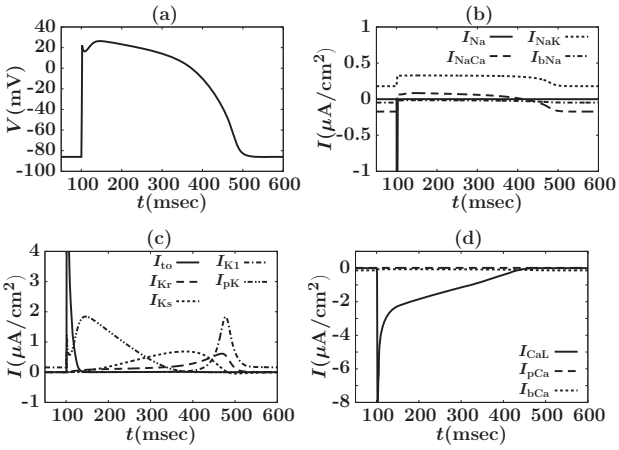

Figure 1: Temporal variation of membrane potential and ionic currents in a standard condition.

$$I_{NaCa} = k_{NaCa} \cdot \frac{e^{\gamma VF/RT}[Na^+]_i^3[Ca^{2+}]_o - e^{(\gamma-1)VF/RT}[Na^+]_o^3[Ca^{2+}]_i \cdot 2.5}{(K_{mNai}^3 + [Na^+]_o^3)(K_{mCa} + [Ca]_o)(1 + k_{sat}e^{(\gamma-1)VF/RT})} \quad (8)$$

$$I_{NaK} = \frac{P_{NaK}[K^+]_o[Na^+]_i}{([K^+]_o + K_{mK})([Na^+]_i + K_{mNa})} \cdot \frac{1}{(1 + 0.1245e^{-0.1VF/RT} + 0.0353e^{-VF/RT})} \quad (9)$$

$$I_{pCa} = c_{pCa}G_{pCa}\frac{[Ca^{2+}]_i}{K_{pCa} + [Ca^{2+}]_i} \quad (10)$$

$$I_{pK} = c_{pK}G_{pK}\frac{V - E_K}{1 + e^{(25-V)/5.98}} \quad (11)$$

$$I_{bNa} = c_{bNa}G_{bNa}(V - E_{Na}) \quad (12)$$

$$I_{bCa} = c_{bCa}G_{bCa}(V - E_{Ca}) \quad (13)$$

where G_{Na}, G_{CaL}, G_{to}, G_{Kr}, G_{Ks}, G_{K1}, G_{pCa}, G_{pK}, G_{bNa} and G_{bCa} (mS/cm^2) are the maximum conductances of ion channels. For the simplicity of bifurcation analyses, we have introduced the coefficients of the maximum conductances c_{Na}, c_{CaL}, c_{to}, c_{Kr}, c_{Ks}, c_{K1}, c_{pCa}, c_{pK}, c_{bNa} and c_{bCa} whose standard values are 1.0. m, h, j, f_1, f_2, x_{r1}, x_s and s are the gating variables, which express opening and closing dynamics of ion channels. Temporal variations of gating variables are described by

$$\frac{d\chi}{dt} = \frac{1}{\tau_\chi}(\chi_\infty - \chi), \quad (\chi = m, h, j, f_1, f_2, x_{r1}, x_s, s) \quad (14)$$

where τ_χ and χ_∞ are the time constant and the steady-state value of the gating variable, respectively. For more details, see the reference [6].

Figure 1 shows the waveforms of membrane potential and ionic currents in a standard condition. The peaks of I_{Na}, I_{to} and I_{CaL} are about -420 μA/cm^2, 18 μA/cm^2 and -13 μA/cm^2, which are omitted in Fig. 1. When the ventricular cells receive the external electrical signals, the inward current (denoted by negative values) I_{Na} is increased sharply and contributes to the upstroke of the membrane potential (depolarization). Then, the outward currents (denoted by positive values), mainly K^+ currents, are activated and the membrane potential is decreased (repolarization).

3. One-Parameter Bifurcation Analysis

The variation of parameters in nonlinear differential equations induces qualitative changes of solutions: bifurcations. In this section, we reveal the bifurcation structure of the reduced TNNP model and examine the method to initiate the pacemaking activity. This paper uses the bifurcation analysis software AUTO [7] for the analysis of the reduced TNNP model.

3.1. The Inward Rectifier Potassium Current I_{K1}

Figure 2(a) shows the one-parameter bifurcation diagram, where the conductance coefficient of the inward rectifier potassium current c_{K1} is the bifurcation parameter. The mambrane potential V in the steady state was plotted for each value of c_{K1} in the diagram. The solid and broken curves show stable and unstable equilibrium points, respectively. The red curve shows the maximum values of stable periodic solutions. The bifurcation points of Hopf, saddle-node, period-doubling, and homoclinic bifurcations are denoted by HB, SN, PD and HC, respectively. Periods of periodic solutions are also shown in the diagram.

In the standard condition ($c_{K1} = 1.0$), a stable equilibrium point corresponding to the resting potential exists (Fig. 2(e)). The stability of equilibrium points changes at the Hopf bifurcation, and the stable periodic solutions are generated. The periodic solutions change their stabilities at PD1 and PD2, and disappear with infinitely large periods at HC. For each value of c_{K1} between HB and PD1 or between PD2 and HC, a stable periodic solution exists. As c_{K1} is increased, the period of periodic solutions increases and its amplitude also increases. Figures 2(c) and 2(d) show typical waveforms of membrane potentials corresponding to the periodic solutions. In the left side of HB or in the right side of SN1, only equilibrium points exist. In both cases, the membrane potentials converge to the stable equilibrium points, but the values of equilibrium points are much different (Fig. 2(b) and 2(e)).

Figure 2 shows that spontaneous action potentials are generated as the inward rectifier potassium current I_{K1} is decreased from the standard value. This result corresponds to the Miake's physiological experiment [1] on the guinue pig ventricular cells, although Fig. 2(a) was obtained using the model of human ventricular cells.

3.2. Ionic Currents during the Pacemaker Activity

Figure 3(a) shows the waveform of the spontaneous action potentials generated by suppression of I_{K1}, and Figs. 3(b)–3(d) are the waveforms of ionic currents. The value of c_{K1} is 0.01.

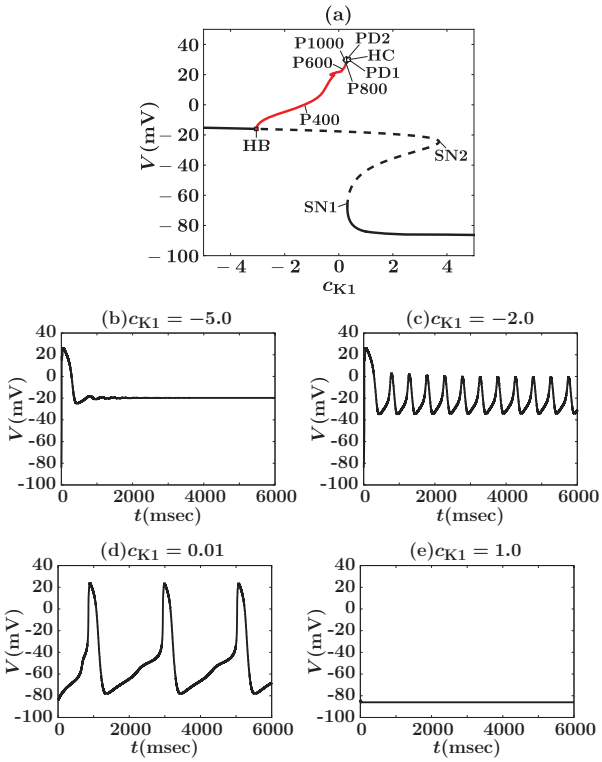

Figure 2: One-parameter bifurcation diagram as for the bifurcation parameter c_{K1}

Pacemaker activity is initiated by slow depolarization generated by I_{bNa}, I_{bCa} (background currents) and I_{NaCa} which exchanges calcium and sodium to maintain the homeostasis of ionic concentration. In intact cells, these inward currents are balanced with the outward I_{K1} current and the resting potential is stable. In the BP cell, absence of balancing I_{K1} causes slow depolarization which leads to the generation of a spontaneous action potential. When V reaches about -60 mV, I_{Na} is activated and accelerates depolarization. In the BP cell, I_{Na} becomes much smaller, which contributes to the fast upstroke in the standard condition. As the membrane potential is increased, inward I_{NaCa} decreases and I_{CaL} is gradually activated, which depolarizes the membrane potential. At the end of slow depolarization, I_{CaL} is fully activated and contributes to the subsequent upstroke and the plateau of the action potential. The potassium currents I_{to}, I_{pK}, I_{Ks}, I_{Kr} are activated in turn and decrease the membrane potential as with the case of the standard condition.

4. Two-Parameter Bifurcation Analysis

The bifurcation points in the one-parameter bifurcation diagram such as Fig. 2(a) are changed by the variation of other parameter values. In this section, we vary two conductance coefficients (c_{K1} and another) simultaneously as the bifurcation parameters to investigate the effects of various ionic currents on the pacemaking activity of the BP cells.

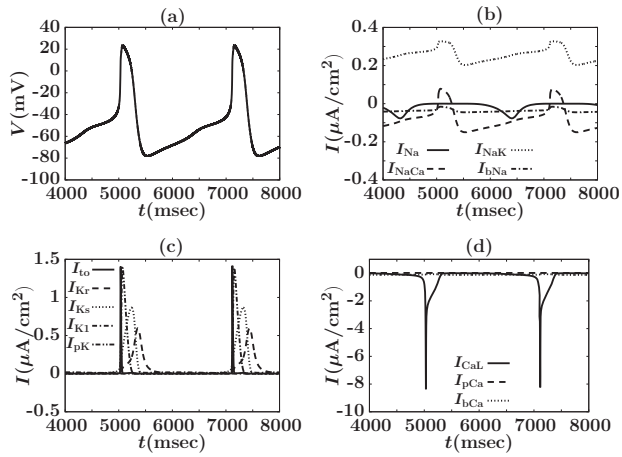

Figure 3: Ionic currents during the pacemaker activity

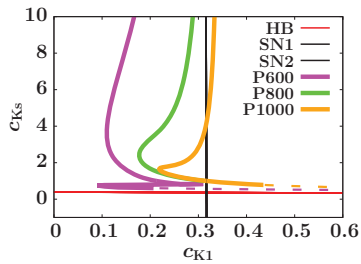

Figure 4: Two-parameter bifurcation diagram as for the two bifurcation parameters c_{K1} and c_{Ks}

4.1. The Inward Rectifier Potassium Current I_{K1} and the Slow Delayed Rectifier Current I_{Ks}

Figure 4 shows the two parameter bifurcation diagram, where c_{K1} and c_{Ks} are the bifurcation parameters. The figure shows the loci of the Hopf bifurcation and the saddle-node bifurcation (bifurcation curves), and the loci of periodic solutions with specific periods which are shown in Fig. 2(a). The loci of periodic solutions represent the contour lines of the periods. The solid and broken contour curves denote stable and unstable periodic orbits, respectively. When c_{Ks} is fixed to 1.0 and c_{K1} is varied, the one-parameter bifurcation diagram Fig. 2(a) can be obtained.

The bifurcation curve of HB is almost parallel to the horizontal axis, although two bifurcation curves of SN are almost parallel to the vertical axis and overlap each other. The periodic solutions exist in the upper side of the HB curve. When c_{Ks} takes the value between 1.0 and 2.5, the periods of periodic solutions become longer as c_{Ks} is increased. As c_{Ks} is further increased, the periods of periodic solutions become shorter, and then, gradually become constant. On the other hand, when c_{Ks} is decreased from the standard value 1.0, the contour lines of the periods approach each other and the periodic solutions become unstable. These results show that the variation of the I_{Ks} current can change the period of the spontaneous pacemaking of the BP. A similar diagram have been obtained by the two-parameter bifurcation analysis as for c_{K1} and c_{Kr}, which is not shown in this paper.

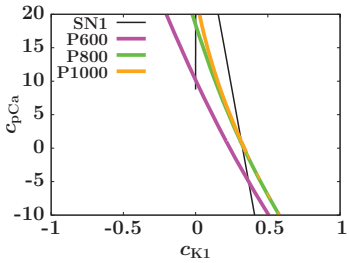

Figure 5: Two-parameter bifurcation diagram as for the two bifurcation parameters c_{K1} and c_{pCa}

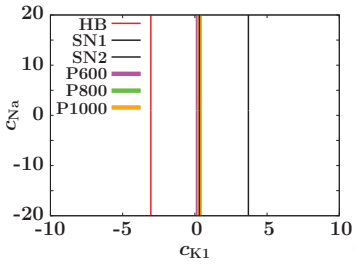

Figure 6: Two-parameter bifurcation diagram as for the two bifurcation parameters c_{K1} and c_{Na}

4.2. The Inward Rectifier Potassium Current I_{K1} and the Plateau Calcium Current I_{pCa}

Figure 5 is the two-parameter bifurcation diagram as for two bifurcation parameters c_{K1} and c_{pCa}. The contour curves of the periods run to the upper left and diverge each other as c_{pCa} is increased. The periods of periodic solutions become longer while c_{pCa} is increased from the standard value. This shows the variation of I_{pCa} has also strong effects of changing periods on the pacemaker activity of the BP.

4.3. The Inward Rectifier Potassium Current I_{K1} and the Fast Sodium Current I_{Na}

The two-parameter bifurcation diagram as for the parameters c_{K1} and c_{Na} is shown in Fig. 6. The bifurcation points and the period of periodic solutions change little as c_{Na} is varied. We have also examined another two-parameter bifurcation diagram, where the bifurcation parameters are c_{K1} and c_{to}. The bifurcation points and the period did not change by the variation of c_{to}, either. This shows I_{Na} and I_{to} have little effect on the period of the spontaneous pacemaking of the BP.

5. Conclusion

We have investigated the method to create the biological pacemaker based on the bifurcation analysis of the reduced TNNP model, which is a human ventricular cell model.

At first, we have examined the one-parameter bifurcation structure, changing the conductance coefficients of ionic currents as the bifurcation parameters. It has been confirmed that the pacemaker activity is induced by the suppression of the inward rectifier potassium current I_{K1}. The spontaneous action potential is initiated by the slow depolarization generated by inward currents I_{NaCa}, I_{bNa} and I_{bCa}. This result corresponds to the preceding studies on the BPs using the guinea pig ventricular cells.

Second, we have varied the two conductance coefficients simultaneously as the bifurcation parameters to investigate the effects of ionic currents on the spontaneous pacemaking. The periods of the spontaneous action potentials induced by the suppression of I_{K1} can be changed by the variation of other ionic currents I_{Ks}, I_{Kr}, I_{pCa}. On the other hand, I_{Na} and I_{to} have little effect on the pacemaking of the BP.

This study gives more detailed and quantitative perspective on the creation of a human BP cell by using a mathematical cell model. Since the reduced TNNP model considers a simplified situation where some variables are fixed to be constant, it is necessary to analyze more detailed models and compare with these results as a future work.

Acknowledgments

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