Role of the Late Sodium Current in Arrhythmias related to Low Repolarization Reserve

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Abstract

The prolongation of action potential duration (APD) is usually related to conditions of low repolarization reserve and leads to long QT syndrome. In these situations, an unbalance between currents can fire early after depolarizations (EADs). This repolarizing disorder has been observed in heart failure situations, where the late sodium current (I_{NaL}) has an important role. In this work we evaluate the effects of I_{NaL} enhancement in the ventricular wall under normal conditions and we analyze the role of I_{NaL} under pathological conditions prone to EADs generation. Human I_{NaL} was formulated and introduced in ten Tusscher AP model. Our results show that the increase in the maximum conductance of I_{NaL} prolongs APD in a rate-dependent manner especially in M cells. A 10-fold increase of I_{NaL} prolongs APD in 35 %, 44 % and 80 % for a stimulation rate of 1 Hz in epicardium, endocardium and M cells, respectively. Finally, the enhancement of I_{NaL} under conditions of low repolarization reserve led to EADs formation in M cells.

1. Introduction

QT interval prolongation is usually related to a delay in phase 3 repolarization, and may have arrhythmogenic consequences in the ventricles (1;2), such as the formation of early afterdepolarizations (EADs) and *Torsade de Pointes* (TdP).

EADs are membrane potential oscillations that occur during the action potential (AP) plateau and may trigger ventricular arrhythmias. An action potential with a long duration increases intracellular calcium (Ca^{2+}) concentration that promotes membrane potential oscillations and formation of EADs, which are also responsible for dispersion of repolarization. These electrical alterations can degenerate into TdP.

On the other hand, pure class III antiarrhythmics drugs (I_{Kr} blockers) can instigate excessive QT interval prolongation due to block of action potential repolarizing currents (3;4). I_{Kr} blockers prolong the APD in a frequency reverse dependent manner and may induce EADs in the ventricle, and even TdP. Drug-induced TdP remains an important problem, especially in combination with risk factors such as bradycardia, hypokalemia or congenital and long QT syndrome. This repolarizing disorder has been observed under pathological situations, such as heart failure (HF), oxidative stress, ventricular hypertrophy and provoke changes in the AP properties and alterations in the functional expression of depolarizing and repolarizing currents. The most relevant changes are down-regulation of potassium (K^+) currents, (repolarization K^+ ionic currents), changes in calcium (Ca^{2+}) handling, decrease of sodium–potassium pump (I_{NaK}) activity and increase of I_{NaL} (5).

The late sodium current (I_{NaL}) is one of the most important currents during repolarization under pathological conditions (6). Several studies have suggested that the I_{NaL} is increased in heart failure and contributes to action potential prolongation and EADs formation (2;7-9). I_{NaL} contributes, at least, to two established HF mechanisms: electrophysiological alterations (interruption of repolarization) and altered cell sodium (Na^+) and Ca^{2+} cycling (10;11). Several authors have hypothesized that an increase of I_{NaL} would potentiate and unmask the proarrhythmic effects of QT prolonging drugs, including drugs that have a very low risk of causing VT (12;13).

The main goal of this study is to evaluate the effects of I_{NaL} enhancement in the different cells of the ventricular wall under normal conditions in a human ventricular AP model. Moreover, we analyze the role of I_{NaL} under pathological conditions prone to EADs generation to shed light the role of late sodium current under these conditions.

2. Methods

In this work, the late sodium current was formulated as an individual ionic current. I_{NaL} was modeled previously by Hund and coworkers (14) for dog ventricular cells, using Hodgkin Huxley formalism (see equations 1 to 4).

$$I_{NaL} = \bar{g}_{NaL} \cdot m_L^{\,3} \cdot h_L \cdot \left(V - E_{NaL}\right) \quad (1)$$

$$\alpha_{m,L} = \frac{0.32 \cdot \left(V_m + 47.13\right)}{1 - e^{\left(-0.1 \cdot \left(V_m + 47.13\right)\right)}} \quad (2)$$

$$\beta_{m,L} = 0.08e^{\left(-V_m/11\right)} \quad (3)$$

$$h_{L,\infty} = \frac{1}{1-e^{\left((V_m+91)/6.1\right)}} \quad (4)$$

Time constant of inactivation (τ_h) and the maximum conductance (g_{NaL}) were modified to reproduce experimental data taken from human (15). The g_{NaL} was fitted to data provided by Tsurugi *et al.* (16) and Maltsev *et al.* (17). In their experiments, they measured an I_{NaL}/I_{NaF} ratio of 0.5% approximately, and g_{NaL} was fitted in our model accordingly. The conductance yielded 0.055 mS/μF. It is to be noted that the ratio does not change with temperature (18). Moreover, experimental data demonstrated that the time constant of inactivation is modified by temperature, for this reason, we multiplied τ_h by the factor Q10 (2.2), so τ_h yielded 233 ms at 37ºC (7;18). Next, we introduced the model of I_{NaL} into the human action potential model formulated by ten Tussher *et al.* (TP06) (19). The TP06 includes the formulation of membrane ionic currents following Hodgkin-Huxley formalism, ionic pumps and exchangers that regulate ionic concentration changes, as well as dynamic changes of intracellular calcium.

To measure the APD_{90} and its rate-dependence, we paced continuously with a basic train of pulses (BCL) of 2 ms in duration and 1.5 times diastolic threshold in amplitude. In all measurements obtained the steady-state was achieved and in the three types of cells: epicardium (epi); endocardium (endo) and M cells. The BCL was modified to observe the APD rate-dependency with I_{NaL} enhancement. Next, we calculated the substration between APD for the maximum and minimum BCL simulated for M cells (APD_{Max} (4000 ms) - APD_{Min} (666 ms)). APD_{90} is assessed as the time between the maximal AP upstroke (dV/dt_{max}) and 90% repolarization from peak amplitude (peak potential minus resting potential).

To analyze the arrhythmogenic role of I_{NaL}, pathological conditions of the cell prone to EAD generation, such as heart failure, ventricular hypertrophy or oxidative stress, were modeled and combined with I_{Kr} block, mimicking the effects of class III antiarrhythmic agents. Specifically, L-type calcium current (I_{CaL}) was 2.4-fold enhanced as well as I_{NaL}. In these simulations, the cell was paced with a BCL of 2000 ms.

3. Results and Discussion

Firstly, we incorporated the I_{NaL} in the TP06 human AP model. APD was prolonged when the current was enhanced, as expected and demonstrated experimentally (20;21). Figure 1 shows an AP when the different types of cells were stimulated with a BCL of 1000 ms. APD_{90} underwent an increase of 7.3 %; 7.4 % and 42 %, when I_{NaL} was included in epi; endo and M cells, respectively. When we increased 10-fold I_{NaL}, APD was prolonged in 44 %, 45 % and 80 %, respectively.

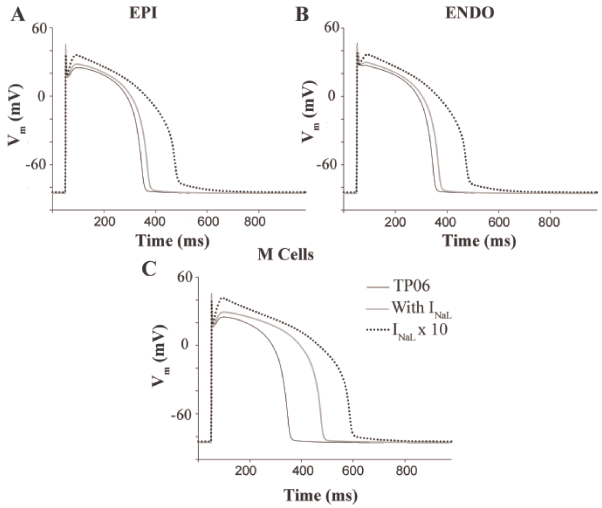

Figure 1. Effects of I_{NaL} on the action potential duration using TP06 (19) (black line), modified TP06 incorporating I_{NaL} (gray line) and 10-fold increased I_{NaL} (dotted line). Panel A) Epicardium; B) endocardium and C) M cells.

The APD rate-dependence in the different types of cells in presence of I_{NaL} is represented in Figure 2. It can be observed that the APD rate-dependence was more pronounced in M cells than epi and endo cells. The raise in APD difference was 666 % ms versus 228 % ms and 360 % ms in epi and endo, respectively, when I_{NaL} was enhanced with respect with TP06, as indicated in Table 1. For BCLs higher than 2000 ms the APD was the same to epi and endo cells. Additionally, we show the prolongation in APD in all BCL values studied, and especially at high BCLs (see Figure 2). Experimentally, Wu *et al.* also observed the rate-dependence, as the increase of APD was significantly higher at longer BCLs (22). Milberg and co-workers observed how veratridine, an activator of I_{NaL}, had more important effects at higher BCLs (8).

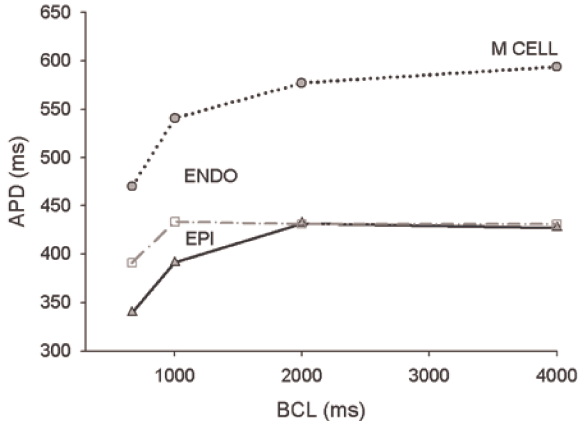

Figure. 2 Influence of the stimulation rate on APD_{90} in epicardium (black line), endocardium (dashed line) and M cells (dotted) when I_{NaL} was enhancement 10-fold.

Table 1. APD difference between BCL of 4000 ms (maximum) and BCL of 666 ms (minimum BCL for M cells).

	EPI	ENDO	M cells
TP06	17,8	19	18
With I_{NaL}	22	28,2	70,9
I_{NaL} x 10	39,5	87,7	123,3

In order to simulate cellular conditions prone to EADs formation and characterize the role of I_{NaL} in the firing of such AP irregularities, some ionic currents were altered. Specifically, a 2.4-fold increase of I_{CaL} was considered, mimicking adrenergic stimulation observed in cases of HF (5) or oxidative stress, as well as the increase of I_{NaL} (15).

In this study, we tested the action of enhancement I_{NaL} (10-fold) combined with a complete block of I_{Kr} under conditions of low repolarization reserve in the diferrent types of cells (endo, epi, M cells). Figure 4 shows alternant EADs under conditions of enhanced I_{NaL} (solid line), highlighting the proarrhythmic effects of this current.

In the case of increased I_{NaL}, EADs arose alternatively and Ca^{2+} overload seemed to be the major responsible for this repolarization irregularity. Intracellular Ca^{2+} overload due to long APD has been implicated as a trigger of spontaneous Ca^{2+} release from sarcoplasmic reticulum causing membrane potential oscillation via Ca^{2+} sensitive currents. In our simulations, we found that EADs arose more easily in M cells than epi and/or endo. The mechanisms for EADs generation have been suggested by the experiments of Choi *et al.* (23), who found how in M cells the primary of EADs was Ca^{2+} overload. Our study supports this observation and provides a powerful tool to get more understanding on the underlying mechanisms. Furthermore, we found that the increase in I_{NaL} favored the initiation of this mechanism.

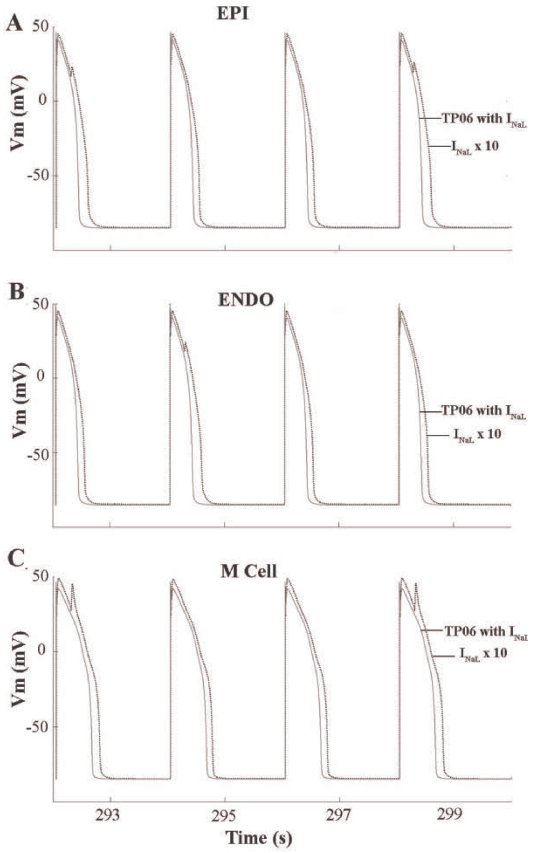

Figure 3. Effects of I_{NaL} enhancement (10–fold) in EADs generation using TP06 (solid line) and modified TP06 with enhanced I_{NaL} (dotted line). Panel A) Epicardium, B) endocardium, C) M cells.

4. Conclusion

In this work, we formulated I_{NaL} for human ventricular myocyte and showed its determinant role in the three different types of cells. Also, we found how I_{NaL} enhancement is proarrhythmic especially in M cells under pathologic conditions prone to EAD generation in the presence I_{Kr} blockers. Finally, our study suggests that targeting I_{NaL} block would have antiarrhythmic effects.

Acknowledgements

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Reference

[1] Volders PG, Vos MA, Szabo B, Sipido KR, de Groot SH, Gorgels AP, et al. Progress in the understanding of cardiac early afterdepolarizations and torsades de pointes: time to revise current concepts. Cardiovasc Res 2000 Jun;46(3):376-92.

[2] Vos MA, Gorenek B, Verduyn SC, van der Hulst FF, Leunissen JD, Dohmen L, et al. Observations on the onset of torsade de pointes arrhythmias in the acquired long QT syndrome. Cardiovasc Res 2000 Dec;48(3):421-9.

[3] Restivo M, Caref EB, Kozhevnikov DO, el Sherif N. Spatial dispersion of repolarization is a key factor in the arrhythmogenicity of long QT syndrome. J Cardiovasc Electrophysiol 2004 Mar;15(3):323-31.

[4] Belardinelli L, Antzelevitch C, Freedman AR. Inhibition of late sodium current: a potnetial drug target to reduce intracellular sodium-dependent calcium overload and its detrimental effects on cardiomyocyte function. S, 13-17. 2004.

[5] Tomaselli GF, Marban E. Electrophysiological remodeling in hypertrophy and heart failure. Cardiovasc Res 1999 May;42(2):270-83.

[6] Maltsev VA, Silverman N, Sabbah HN, Undrovinas AI. Chronic heart failure slows late sodium current in human and canine ventricular myocytes: implications for repolarization variability. Eur J Heart Fail 2007 Mar;9(3):219-27.

[7] Maltsev VA, Sabbah HN, Higgins RS, Silverman N, Lesch M, Undrovinas AI. Novel, ultraslow inactivating sodium current in human ventricular cardiomyocytes. Circulation 1998 Dec 8;98(23):2545-52.

[8] Milberg P, Pott C, Fink M, Frommeyer G, Matsuda T, Baba A, et al. Inhibition of the Na+/Ca2+ exchanger suppresses torsades de pointes in an intact heart model of long QT syndrome-2 and long QT syndrome-3. Heart Rhythm 2008 Oct;5(10):1444-52.

[9] Zaza A, Belardinelli L, Shryock JC. Pathophysiology and pharmacology of the cardiac "late sodium current.". Pharmacol Ther 2008 Sep;119(3):326-39.

[10] Zygmunt AC, Eddlestone GT, Thomas GP, Nesterenko VV, Antzelevitch C. Larger late sodium conductance in M cells contributes to electrical heterogeneity in canine ventricle. Am J Physiol Heart Circ Physiol 2001 Aug;281(2):H689-H697.

[11] Song Y, Shryock JC, Wu L, Belardinelli L. Antagonism by ranolazine of the pro-arrhythmic effects of increasing late INa in guinea pig ventricular myocytes. J Cardiovasc Pharmacol 2004 Aug;44(2):192-9.

[12] Noble D, Noble PJ. Late sodium current in the pathophysiology of cardiovascular disease: consequences of sodium-calcium overload. Heart 2006 Jul;92 Suppl 4:iv1-iv5.

[13] Orth PM, Hesketh JC, Mak CK, Yang Y, Lin S, Beatch GN, et al. RSD1235 blocks late INa and suppresses early afterdepolarizations and torsades de pointes induced by class III agents. Cardiovasc Res 2006 Jun 1;70(3):486-96.

[14] Hund TJ, Rudy Y. Rate dependence and regulation of action potential and calcium transient in a canine cardiac ventricular cell model. Circulation 2004 Nov 16;110(20):3168-74.

[15] Maltsev VA, Sabbah HN, Undrovinas AI. Late sodium current is a novel target for amiodarone: studies in failing human myocardium. J Mol Cell Cardiol 2001 May;33(5):923-32.

[16] Tsurugi T, Nagatomo T, Abe H, Oginosawa Y, Takemasa H, Kohno R, et al. Differential modulation of late sodium current by protein kinase A in R1623Q mutant of LQT3. Life Sci 2009 Mar 13;84(11-12):380-7.

[17] Maltsev VA, Kyle JW, Undrovinas A. Late Na+ current produced by human cardiac Na+ channel isoform Nav1.5 is modulated by its beta1 subunit. J Physiol Sci 2009 May;59(3):217-25.

[18] Nagatomo T, Fan Z, Ye B, Tonkovich GS, January CT, Kyle JW, et al. Temperature dependence of early and late currents in human cardiac wild-type and long Q-T DeltaKPQ Na+ channels. Am J Physiol 1998 Dec;275(6 Pt 2):H2016-H2024.

[19] Ten Tusscher KH, Bernus O, Hren R, Panfilov AV. Comparison of electrophysiological models for human ventricular cells and tissues. Prog Biophys Mol Biol 2006 Jan;90(1-3):326-45.

[20] Drouin E, Charpentier F, Gauthier C, Laurent K, Le Marec H. Electrophysiologic characteristics of cells spanning the left ventricular wall of human heart: evidence for presence of M cells. J Am Coll Cardiol 1995 Jul;26(1):185-92.

[21] Li GR, Feng J, Yue L, Carrier M. Transmural heterogeneity of action potentials and Ito1 in myocytes isolated from the human right ventricle. Am J Physiol 1998 Aug;275(2 Pt 2):H369-H377.

[22] Wu L, Shryock JC, Song Y, Li Y, Antzelevitch C, Belardinelli L. Antiarrhythmic effects of ranolazine in a guinea pig in vitro model of long-QT syndrome. J Pharmacol Exp Ther 2004 Aug;310(2):599-605.

[23] Choi BR, Burton F, Salama G. Cytosolic Ca2+ triggers early afterdepolarizations and Torsade de Pointes in rabbit hearts with type 2 long QT syndrome. J Physiol 2002 Sep 1;543(Pt 2):615-31.

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