Late sodium current and cardiac electrical and mechanical dysfunction

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Abstract

Late sodium current ($I_{Na,L}$) is a small sustained current existed during repolarization phase of cardiac action potential after peak I_{Na} is complete. Although relatively small in magnitude in normal heart (endogenous late I_{Na}), it represents a functional relevant contributor to cardiomyocyte repolarization and is potentially proarrhythmic in hearts with reduced repolarization reserve. The increased amplitude of late I_{Na} (enhanced late I_{Na}) is recognized in patients with long QT syndrome 3 (LQTS 3), as well as plays an important role in the arrhythmogenesis and mechanical dysfunction under various cardiovascular pathological and pharmacological conditions, including slow heart rate, myocardial ischemia and reperfusion injury and heart failure. Basic and clinical research indicates that inhibition of late I_{Na} is effective in preventing and treating cardiac arrhythmias with different mechanisms, and improving ventricular pump (?) function. Selective inhibitor of late sodium current has no or minimal effect on peak sodium current and/or I_{Kr}, which may have no or minimal risk of proarrhythmia in comparing with classic class I or III antiarrhythmic drugs (AADs), especially in patients with ischemic heart diseases. With the increase in understanding of the late sodium current, it may be helpful for clinicians to use late sodium current inhibitors to treat cardiac arrhythmias in patients with the enhancement of late sodium current, such as LQT 3, etc. In this review, we will discuss the mechanistic role of endogenous and enhanced late sodium current in arrhythmogenesis and mechanical dysfunction, and the late sodium current targeted applications.

This review illustrates a wide range of situations that may accentuate $I_{Na\text{-}L}$. These include (1) overlaps between steady-state activation and inactivation increasing *window current*, (2) kinetic deficiencies in Na^+ channel inactivation leading to *bursting phenomena* associated with repetitive channel openings and (3) *non-equilibrium gating* processes causing channel re-opening due to more rapid recoveries from inactivation. All these biophysical possibilities were identified in a selection of abnormal human *SCN5A* genotypes. The latter presented as a broad range of clinical arrhythmic phenotypes, for which effective therapeutic intervention

would require specific identification and targeting of the diverse electrophysiological abnormalities underlying their increased $I_{Na\text{-}L}$.

Introduction

. The normal function of cardiomyocytes relays in part on the coordinate work of various ion channels acrossing sarcoplasmic membrane of these cells including voltage gated Na, potassium and Ca^{2+} ion channels. The voltage-gated sodium channel Nav1.5, encoded by gene of SCN5A, is primarily expressed in cardiaomyocytes, where it mediates the influx of Na^+ across cell membrane. Depolarization causes transient (1~3 ms) opening of sodium channels, followed by a large amount of Na^+, following the concentration gradient, rushing into cells to further depolarize them and generating the upstroke of action potentials (APs). The transient but large sodium current (I_{Na}) during this period is termed as peak I_{Na}. While the majority of these channels inactivate quickly and remain inactivated until the repolarization of cell membrane is complete,however, there are still some channels remaining opened or reopen after inactivation, allowing influx of Na^+ during repolarization period and resulting in a tiny late component of sodium current (late $I_{Na)}$ that may persist for several hundred of milliseconds throughout the repolarization of an AP. The magnitude of late I_{Na} is small (<0.5 % of peak I_{Na})[1-3] which cause no significant changes in the duration and configuration of an AP without any change in mechanical function of the cells (Fig. 1). Therefore, late I_{Na} was seen just as the sustained tail of sodium current for many years until the identification of mutations in the gene SCN5A that caused the congenital long-QT syndrome type 3[4]. In recent years, it attracts increasing interests because enhanced late I_{Na} is associated with many pathological, pharmacological and toxicological conditions and may cause an increase in the duration of an AP (APD) with proarrhythmic activities, i.e., EAD and DAD, (Fig. 1) [5, 6]. In the last decade, increasing evidence indicates that late I_{Na} could be a new and promising therapeutic target for the treatment of cardiac ischemia and cardiac arrhythmias [7-10]. Clinical evidence indicates that class I antiarrhythmic drugs inhibit peak I_{Na} to slow myocardial conduction and increase the mortality in patients with ischemic heart disease. Because selective late I_{Na} inhibitors affects

neither peak I_{Na} nor I_K at or slightly above therapeutic concentrations, they may have low or no chance to cause proarrhythmic activities (Fig. 2).

The mechanism of endogenous and enhanced late I_{Na}

Although it is small under normal circumstances, endogenous late I_{Na} may be relatively greater in cardiac cells with slow rate and Pukenji cells. The enhanced magnitude of late I_{Na} is seen under many conditions, including gene mutations[4, 11-14], or pharmacological agents, myocardial ischemia and reperfusion[15, 16], heart failure[17, 18], etc. Table 1 in a recent review [19] lists conditions associated with increased late $I_{Na.}$

Sodium channel is a macro-molecular complex consisting of a primary α subunit and other regulatory subunits including β1, β2, β3 and β4 [20]. It also assembles with a variety of other proteins, including Snat1[14], Cav3[21] and CaMK-II[22], to form a complex with sodium channels and influence its function/expression. Thus, mutations of any gene encoding these proteins might cause abnormities of sodium current. For example, gain-of-function mutations in SCN5A disrupt Na^+ inactivation and consequently result in increased I_{Na} presented as LQT3[4]. In addition, gene mutations of CAV3, SCN4B and SNTA1 are responsible for LQT9, LQT10 and LQT12, respectively[13, 14, 23]. Rett syndrome is a neuro developmental disorder caused by mutations in methyl-CpG–binding protein 2 (MECP2). McCauley et al. found that the prolongation of the QTc interval was common in patients with Rett syndrome, and late I_{Na} was greater in cardiomyocytes isolated from both Mecp2Null/Y and nervous system–specific conditional Mecp2 knockout mice[24].

Late I_{Na} is also regulated by endogenous agents such as Ca^{2+}-CaM dependent protein kinase Ⅱ (CaMK Ⅱ), protein kinase C (PKC), nitric oxide (NO) and carbon monoxide (CO), via post-translational modification of the channel protein. These agents modulate sodium channel function individually and/or cooperatively. Ca2+/calmodulin-dependent protein kinase IIδ (CaMK IIδ), the predominant isoform of CaML II expressed in the heart, is the most well-known signaling pathway for the regulation of Nav1.5. CaMKII facilitates late I_{Na} through phosphorylating Thr-594

and Ser-516 of Nav1.5 to enhance accumulation of sodium channel into an intermediate inactivated state[22]. Ser571 is also an important regulatory locus for CaMK II, and its phosphorylation is increased in vitro and human failure hearts [25-27].

Protein kinase C (PKC) is a group of serine–threonine protein kinases, and there are at least seven members identified in mammalian myocardium[28]. Some of them are calcium-dependent and others that are insensitive to Ca^{2+} could be activated by diacylglycerol (DAG) or lipid-derived second messengers[29]. The phosphorylation of Nav1.5 at S1503 is involved in the inactivation of sodium channels. Therefore, inhibition of PKC causes an increase in late I_{Na}[30-33] .

Late I_{Na} is also affected by other endogenous agents or cellular metabolites. PKA is found to increase late I_{Na} in disease-linked sodium channel mutant (D1790G)[34]. H_2O_2 and free radicals were found to stimulate late I_{Na}, and inhibition of late I_{Na} by ranolazine or TTX attenuated the AP lengthening by H_2O_2[35]. This effect may come from the free radicals' directly activation on CaMK II[36]. Lysophosphatidylcholine (LPC) accumulates under ischemic conditions, and it may increase cardiac late I_{Na} through peroxynitrite formation[15]. Nitric oxide (NO) could reduce the inactivation of Na^+ channels, thus increase late I_{Na}[37].

In addition, sodium current is modulated under pathological conditions which ectopically active signaling pathways or accumulate cellular metabolites. Increased late I_{Na} in ischemia may be caused by acidosis and ischemic metabolites such as LPC as indicated above. Calcium overloading occurs in heart failure, which excessively activates CaMK II resulting in augmentation of late I_{Na}.

In summary, these pathological conditions and agents augment late I_{Na} by disrupting the inactivation of sodium channels, turning sodium channel into an intermediate inactivated state that is susceptible to be re-opened or fast recovery of channels from inactivation during non-equilibrium conditions[10].

Late I_{Na} associated arrhythmias and mechanical dysfunction

Late I_{Na} associated arrhythmias

For its small magnitude, the effects of late I_{Na} on AP and arrhythmia are under debate for a long time. It should be noticed that the net current flowing during AP plateau is relative small and the algebraic sum of the currents for repolarization is close to zero during this phase. Therefore, even subtle changes in any inward or outward current can have a relative larger impact on AP morphology.

It has been observed that TTX shortened the APD and an increase in late I_{Na} lengthened APD[3, 38]. In our previous studies, inhibition of outward potassium current would lengthen APD of rabbit cardiomyocytes, increase transmural dispersion of repolarization, and result in torsade de pointes ventricular tachycardia. These effects were more obvious at low pacing rate, i.e., reverse rate dependent. While application of inhibitor that decreases endogenous late I_{Na} could attenuate these effects [39, 40].As mentioned above, late I_{Na} is increased under many pathological or pharmacological conditions. Enhanced late I_{Na} may initiate and maintain arrhythmia through inducing after-polarization (early and delayed), and reentry[10].

During the AP plateau, either an increase in inward currents (late I_{Na}, $I_{Ca\text{-}L}$) or a decrease in outward repolarizing currents (I_{Kr}, I_{Ks}) prolongs the duration of AP plateau, corresponding with a lengthened QT interval in ECG recording. The prolonged AP plateau facilitates the occurrence of early after-depolarizations (EADs), a primary arrhythmic activity. Inward currents, including $I_{Ca\text{-}L}$[41], sodium current, and forward mode of the sodium/calcium exchange current (I_{NCX}) are responsible for the upstroke of an EAD[38]. Increased late I_{Na} is associated with AP prolongation, which provide adequate time for L-type Ca^{2+} channels to recover from inactivation. In addition, increased intracellular Ca^{2+} drives a reverse mode of I_{NCX}, generating a depolarizing current[42]. All these currents contribute to the formation of an EAD.

Delayed after-depolarizations (DADs) are transient depolarizations of the cell membrane following repolarization of a previous AP, and the transient inward current (I_{Ti}), which is observed under condition of overloaded intracellular Ca^{2+}, is responsible for a DAD[19]. Increasing evidence indicates that enhanced late I_{Na} is associated with DADs, and inhibition of late I_{Na} suppresses DADs and other trigged activities[43, 44]. The underlying mechanisms may be that increased intracellular Na^{+}

following enhanced late I_{Na} decreases the Na^+ gradient to extrude Ca^{2+} through Na–Ca exchange, and even promotes Na–Ca exchanger to work in a reverse mode with Ca^{2+} actually entering the cell[45].

Normal transmural heterogeneity of an AP in the heart may act as substrate for reentrant arrhythmia in certain conditions. Transmural differences in ionic currents densities underlie these heterogeneities. Differences in the density of late I_{Na} are also observed in canine and murine hearts [46, 47]. In the ventricles, late I_{Na} was found to be larger in M cells and Purkinje fiber than in the epicardial or endocardial cells[48]. Mutations causing facilitation of late I_{Na} are found to be associated with increased QT dispersion, this may partly be explained by the difference in the dispersion of distribution of the late I_{Na} [49]. Enhanced late I_{Na} increases transmural dispersion of repolarization time and this effect could be suppressed by inhibiting late I_{Na} [50].

LQT3 results from the "gain-of-function" mutations of SCN5A encoding the α subunit of sodium current. The first reported LQT3 mutation in patient was ΔKPQ, a deletion of amino acids 1505–1507 in NaV1.5[4]. Since then, more than 80 SCN5A mutations associated with an increase in late I_{Na} have been reported in patients with LQT3. Most of these mutations increase sodium current by delaying Na^+ channel to inactivate or facilitating it recover from the inactivation during the plateau of an AP and reopen, thereby rendering the inactivation process to be unstable or slow or incomplete in association with increasing number of open channels.

Dysfunction of other proteins assembled in sodium channel complex may also enhance late I_{Na}, and thus lead to LQT. Caveolin 3, an integral membrane protein encoded by CAV3, plays important roles in signal transduction and vesicular transportation. In 2006, Vatta, et al, found two kinds mutation of CAV3 (F97C and S141R) in LQT patients which resulted in a 2- to 3- fold increase in late I_{Na}[23]. Cheng, et al, demonstrated that the F97C CAV3 mutation relieved the repressive effect of caveolin on NO synthase 1, which increased S-nitrosylation of SCN5A and late I_{Na}[21]. Sodium channel has a main pore-performing α subunit and is associate with one or more auxiliary β subunits. In human, four different β subunits have been identified and perturbation of any of these components may affect the normal function

of sodium channel. Medeiros-Domingo, et al, reproted a LQT pedigree with L179F-SCN4B mutation[13]. Heterologous expression of this mutated subunit caused an 8-fold increase in late I_{Na}, and this new type of LQT was classified as LQT10. Another type of LQT associated with increased late I_{Na} is LQT12, which was induced by mutation of α-1-syntrophin. Encoded by SNTA1, α-1-syntrophin interacts with SCN5A, nNOS, and PMCA4b, while A390V-SNTA1 disrupted interact of PMCA4b with this complex and increases nitrosylation of SCN5A, leads to an increase in both peak and late I_{Na} [14].

Late I_{Na} associated mechanical dysfunction

Na^+ enters the cell through I_{Na}, Na–Ca exchange and Na–H exchange, and leaves the cell through Na–K ATPase. The amount of Na^+ enters the cell is called "Na^+ loading". Na^+ loading is determined by the amplitude and duration of sodium current [51]. Although the amplitude of late I_{Na} is much lower than peak I_{Na} (<0.5 %) under normal circumstances, it contributes the majority of Na^+ loading because the duration is much longer, especially under pathological conditions, e.g. myocardial ischemia, heart failure[19]. The increase in the amplitude of late I_{Na} results in an extension of the duration of phase 2 of an AP. The increase of the amplitude of late I_{Na} and the duration of phase 2 of an AP leads to increases in Na^+ loading and intracellular Na^+ concentration. Increased intracellular Na^+ concentration will cause an increase in intracellular Ca^{2+} concentration by reverse mode of NCX [45]. The direct effect of increased Ca^{2+} concentration on mechanical function is impaired relaxation or decreased diastolic function. The increase of late I_{Na} and subsequent Ca^{2+} overloading cause an elevation of ventricular end-diastolic pressure[45]. With increased diastolic tone, it also increases the resistance to blood flow in the microcirculation resulting in reduced oxygen supply. A vicious cycle is thus created where diastolic dysfunction after ischemia and heart failure increases oxygen and energy consumption and aggravates deficiency of oxygen in ischemic myocardial tissue and failing myocardium [52] (Fig. 3).

Therapeutic applications of late I_{Na} inhibitors

Because increased late I_{Na} causes arrhythmias and mechanical dysfunction, it is an attractive target for the treatment. There have been some studies demonstrated that inhibition of late I_{Na} play a therapeutic role on arrhythmias, myocardial inchemia and mechanical dysfunction.

Therapeutic applications in cardiac arrhythmias

Basic studies have demonstrated that late I_{Na} inhibitors suppress ventricular arrhythmias (e.g. torsade de pointes VT and VF)[39, 40, 53-57]. In hearts with ventricular arrhythmias, the inhibition of late I_{Na} reduces repolarization heterogeneity due to the preferential abbreviation of an AP of M cell and suppresses EAD- and DAD- triggered activity[58-61]. Inhibition of endogenous late I_{Na} attenuated MAPD prolongation, beat-to-beat variability of repolarization (BVR), reverse rate dependence (RRD) and abolished arrhythmic activity caused by I_{Kr} inhibitors[39, 40]. When late I_{Na} was increased, inhibitions of either endogenous or augmented late I_{Na} were responsible for the elimination of the proarrhythmic effects of some antiarrhythmic drugs (e.g., amiodarone, quinidine and moxifloxacin, etc.) [55-57]. In addition, Kumar, et al, reported that ranolazine (9.2 +/- 2.1 microM in plasma level) increased ERP and raised repetitive extrasystole threshold, and VF threshold in porcine model[53]. Nieminen, et al, also found that ranolazine increased VF threshold and suppressed ischemia-induced increase in T-wave alternans significantly in severe coronary stenosed porcine hearts[54]. Some class Ia and Ic antiarrhythmic drugs also have the inhibitory effects on late I_{Na} with no or minimal selectivity on peak $I_{Na.}$ In contrast, selective late I_{Na} inhibitors (e.g., ranolazine) have no significant effect on peak I_{Na} at therapeutic concentration range. So they may not induce reentry and proarrhythmic activities when they are used in clinic. Recently, Bacic et al reported that a novel selective late I_{Na} inhibitor eleclazine was effective in treating catecholamine-induced ventricular tachycardias and T-wave atternans in an intact porcine model and the effect was better than flecainide[62].

In clinical studies, inhibition of late I_{Na} is also reported to attenuate ventricular arrhythmias in patients with LQTS. Compared to β blockers in the treatment of LQTS, late I_{Na} inhibitor may be potentially used as a specific, at least in some types of

mutations, the treatment of LQT 3. Moss, et al, reported that ranolazine shortened a prolonged QTc interval in patients with the LQT3-ΔKPQ mutation at therapeutic concentrations[63]. Recently, Mazzanti et al found that another late sodium current inhibitor, mexiletine, caused a major reduction of life-threatening arrhythmic events in LQT3 patients besides shortening QTc interval in a retrospective cohort[64]. Besides LQT3, late sodium current inhibitors also have effect on other LQTS. Gao, et al, treated a LQT8 patient with mexiletine and found that mexiletine shortened QTc, attenuated QT–RR slope, abolished 2:1 AV block and T wave alternans. In MERLIN (metabolic efficiency with ranolazine for less ischemia in non-ST-elevation acute coronary syndrome)-TIMI36 trial, ranolazine was found to reduce ventricular (non sustained ventricular tachycardia) and atial (new atrial fibrillation) arrhythmias in patients with non ST-segment elevated acute coronary syndrome[65].

In animals and humans with atrial fibrillation, APD and ERP are shortened. Ranolazine blocked I_{Kr} and peak I_{Na} to prolong APD and ERP to increase post-repolarization refractoriness and decrease conduction velocity[61]. Because selective late I_{Na} block does not prolong ERP, reduction of late I_{Na} only is not supposed to be significantly effective for the suppression of AF. However, recent studies indicated that inhibition of late I_{Na} by GS-458967 could suppressed atrial fibrillation caused by augmented late I_{Na} in animal models[66, 67].

Therapeutic applications in mechanical dysfunction

Late I_{Na} inhibitors have been reported to decrease sodium and calcium loading, reduce oxidative stress and improve ventricular function [35, 68-73]. Rocchetti, et al, reported that monocrotaline (MCT) induced pulmonary arterial hypertension (PAH), increased right ventricular (RV) systolic pressure, and caused RV hypertrophy. In the RV, collagen was increased, and myocytes were enlarged with T-tubule disarray and displayed myosin heavy chain isoform switch. Late I_{Na} was also markedly enhanced. Ranolazine completely prevented the increase in late I_{Na} and limited PAH-induced remodeling in RV[74]. Rastogi, et al, reported that in dogs with moderate heart failure, compared with the placebo group, ranolazine alone or in

combined with enalapril or metaprolol prevented the increase in end-diastolic volume (EDV) and end-systolic volume (ESV) with a significant increase in ejection fraction (EF), which was associated with beneficial effects at the cellular level on histomorphometric parameters, including hypertrophy, fibrosis, and capillary density as well as the expression for pathological hypertrophy and Ca^{2+} cycling genes[75]. In Dahl salt-sensitive (DSS) rats model, compared with a normal salt diet, 8 weeks high-salt diet caused increases in LV mass and diastolic dysfunction [isovolumic relaxation time (IVRT), early transmitral flow velocity/early mitral annulus velocity (E/E') ratio. Late I_{Na} in LV myocytes from high-salt rats was significantly increased. Selective late I_{Na} inhibitor GS-967 decreased the enhanced late I_{Na}. Chronic treatment with GS-967 reduced the LV mass, increased the E/E' ratio, and prolonged IVRT without affecting blood pressure or LV systolic function[73].

In clinical studies, the MARISA (monotherapy assessment of ranolazine in stable angina) study was a randomized, multicenter, double-blind, dose-finding study utilizing ranolazine as monotherapy for angina. The result indicated that ranolazine increased the exercise duration, exercise time to angina, and exercise time to 1-mm ST segment depression in dose dependent ways after 4 weeks of treatment[76]. In contrast, the CARISA (combination assessment of ranolazine in stable angina) and ERICA (efficacy of ranolazine in chronic angina) trial were performed to evaluate the efficacy of ranolazine on the basis of standard antianginal drugs. In CARISA study, ranolazine significantly increased the time of exercise, the time to onset of angina during exercise and ST-segment depression after 12 weeks of therapy. Angina frequency was also reduced compared to placebo[77]. In ERICA trial, ranolazine significantly reduced the number of weekly angina episodes compared to placebo[78]. Moreover, The results of the TERISA (type 2 diabetes evaluation of ranolazine in subjects with chronic stable angina) trial showed that the average weekly number of angina episodes among diabetic patients was significantly reduced in the ranolazine group compared with placebo[79].

To evaluate the effect of ranolazine on unstable angina, The MERLIN -TIMI36 trial was conducted. Although there was no difference in the rate of cardiovascular death,

myocardial infarction or recurrent ischemia between ranolazine and placebo group. However, in patients with brainnatriuretic peptide (BNP) >80 pg/ml, ranolazine reduced the rate of cardiovascular death, MI, and recurrent ischemia. In addition, the safety of ranolazine in ischemic patients was confirmed in the MERLIN-TIMI 36 trial because of the sample size. The most frequent adverse events were dizziness, nausea and constipation[80]. Khan et al performed a pilot study to investigate the effect of ranolazine on exercise capacity, right ventricular indices, and hemodynamic characteristics in patients with PAH. After 3 months treatment, ranolazine reduced RV size, improved RV function (improvement in RV strain during exercise at 3 months), and made a trend toward improved exercise time and exercise watts on bicycle echocardiography, but was not associated with improvement in invasive hemodynamic parameters[81].

5 Conclusion

Increasing evidence suggests that both endogenous and enhanced late I_{Na} play an important role in the genesis of cardiac arrhythmias and mechanical dysfunction. Compared with other drugs, selective inhibitors of late I_{Na} has no or minimal effect on electrophysiological properties, blood pressure and heart rate. Therefore, late I_{Na} may become a promising therapeutic target for arrhythmia and mechanical dysfunction.

Reference

1 Maltsev VA, Undrovinas AI. A multi-modal composition of the late Na+ current in human ventricular cardiomyocytes. Cardiovasc Res 2006, 69:116-127.

2 Patlak JB, Ortiz M. Slow currents through single sodium channels of the adult rat heart. J Gen Physiol 1985, 86:89-104.

3 Kiyosue T, Arita M. Late sodium current and its contribution to action potential configuration in guinea pig ventricular myocytes. Circ Res 1989, 64:389-397.

4 Wang Q, Shen J, Splawski I et al. SCN5A mutations associated with an inherited cardiac arrhythmia, long QT syndrome. Cell 1995, 80:805-811.

5 Gupta T, Khera S, Kolte D et al. Antiarrhythmic properties of ranolazine: A review of the current evidence. Int J Cardiol 2015, 187:66-74.

6 Sossalla S, Maier LS. Role of ranolazine in angina, heart failure, arrhythmias, and diabetes. Pharmacol Ther 2012, 133:311-323.

7 Zaza A, Belardinelli L, Shryock JC. Pathophysiology and pharmacology of the cardiac "late sodium current.". Pharmacol Ther 2008, 119:326-339.

8 Moreno JD, Clancy CE. Pathophysiology of the cardiac late Na current and its potential as a drug target. J Mol Cell Cardiol 2012, 52:608-619.

9 Zaza A, Rocchetti M. The late Na+ current--origin and pathophysiological relevance. Cardiovasc Drugs Ther 2013, 27:61-68.

10 Shryock JC, Song Y, Rajamani S et al. The arrhythmogenic consequences of increasing late INa in the cardiomyocyte. Cardiovasc Res 2013, 99:600-611.

11 Brintzinger I. [Education to cleanliness for children]. Dtsch Krankenpflegez 1991, 44:190-192.

12 Maguy A, Hebert TE, Nattel S. Involvement of lipid rafts and caveolae in cardiac ion channel function. Cardiovasc Res 2006, 69:798-807.

13 Medeiros-Domingo A, Kaku T, Tester DJ et al. SCN4B-encoded sodium channel beta4 subunit in congenital long-QT syndrome. Circulation 2007, 116:134-142.

14 Ueda K, Valdivia C, Medeiros-Domingo A et al. Syntrophin mutation associated with long QT syndrome through activation of the nNOS-SCN5A macromolecular complex. Proc Natl Acad Sci U S A 2008, 105:9355-9360.

15 Gautier M, Zhang H, Fearon IM. Peroxynitrite formation mediates LPC-induced augmentation of cardiac late sodium currents. J Mol Cell Cardiol 2008, 44:241-251.

16 Trenor B, Gomis-Tena J, Cardona K et al. In silico assessment of drug safety in human heart applied to late sodium current blockers. Channels (Austin) 2013, 7:249-262.

17 Maltsev VA, Silverman N, Sabbah HN et al. Chronic heart failure slows late sodium current in human and canine ventricular myocytes: implications for repolarization variability. Eur J Heart Fail 2007, 9:219-227.

18 Undrovinas AI, Maltsev VA, Sabbah HN. Repolarization abnormalities in cardiomyocytes of dogs with chronic heart failure: role of sustained inward current. Cell Mol Life Sci 1999, 55:494-505.

19 Belardinelli L, Giles WR, Rajamani S et al. Cardiac late Na(+) current: proarrhythmic effects, roles in long QT syndromes, and pathological relationship to CaMKII and oxidative stress. Heart Rhythm 2015, 12:440-448.

20 Isom LL, De Jongh KS, Patton DE et al. Primary structure and functional expression of the beta 1 subunit of the rat brain sodium channel. Science 1992, 256:839-842.

21 Cheng J, Valdivia CR, Vaidyanathan R et al. Caveolin-3 suppresses late sodium current by inhibiting nNOS-dependent S-nitrosylation of SCN5A. J Mol Cell Cardiol 2013, 61:102-110.

22 Ashpole NM, Herren AW, Ginsburg KS et al. Ca2+/calmodulin-dependent protein kinase II (CaMKII) regulates cardiac sodium channel NaV1.5 gating by multiple phosphorylation sites. J Biol Chem 2012, 287:19856-19869.

23 Vatta M, Ackerman MJ, Ye B et al. Mutant caveolin-3 induces persistent late sodium current and is associated with long-QT syndrome. Circulation 2006, 114:2104-2112.

24 McCauley MD, Wang T, Mike E et al. Pathogenesis of lethal cardiac arrhythmias in Mecp2 mutant mice: implication for therapy in Rett syndrome. Sci Transl Med 2011, 3:113ra125.

25 Unudurthi SD, Hund TJ. Late sodium current dysregulation as a causal factor in arrhythmia. Expert Rev Cardiovasc Ther 2016, 14:545-547.

26 Hund TJ, Koval OM, Li J et al. A beta(IV)-spectrin/CaMKII signaling complex is essential for membrane excitability in mice. J Clin Invest 2010, 120:3508-3519.

27 Koval OM, Snyder JS, Wolf RM et al. Ca2+/calmodulin-dependent protein kinase II-based regulation of voltage-gated Na+ channel in cardiac disease. Circulation 2012, 126:2084-2094.

28 Banyasz T, Szentandrassy N, Magyar J et al. An emerging antiarrhythmic target: late sodium current. Curr Pharm Des 2015, 21:1073-1090.

29 Palaniyandi SS, Sun L, Ferreira JC et al. Protein kinase C in heart failure: a therapeutic target? Cardiovasc Res 2009, 82:229-239.

30 Astman N, Gutnick MJ, Fleidervish IA. Activation of protein kinase C increases neuronal excitability by regulating persistent Na+ current in mouse neocortical slices. J Neurophysiol 1998, 80:1547-1551.

31 Ma J, Luo A, Wu L et al. Calmodulin kinase II and protein kinase C mediate the effect of increased intracellular calcium to augment late sodium current in rabbit ventricular myocytes. Am J Physiol Cell Physiol 2012, 302:C1141-1151.

32 West JW, Numann R, Murphy BJ et al. A phosphorylation site in the Na+ channel required for modulation by protein kinase C. Science 1991, 254:866-868.

33 Wu Y, Wang L, Ma J et al. Protein kinase C and Ca(2+) -calmodulin-dependent protein kinase II mediate the enlarged reverse INCX induced by ouabain-increased late sodium current in rabbit ventricular myocytes. Exp Physiol 2015, 100:399-409.

34 Tateyama M, Rivolta I, Clancy CE et al. Modulation of cardiac sodium channel gating by protein kinase A can be altered by disease-linked mutation. J Biol Chem 2003, 278:46718-46726.

35 Song Y, Shryock JC, Wagner S et al. Blocking late sodium current reduces hydrogen peroxide-induced arrhythmogenic activity and contractile dysfunction. J Pharmacol Exp Ther 2006, 318:214-222.

36 Erickson JR, Joiner ML, Guan X et al. A dynamic pathway for calcium-independent activation of CaMKII by methionine oxidation. Cell 2008, 133:462-474.

37 Ahern GP, Hsu SF, Klyachko VA et al. Induction of persistent sodium current by exogenous and endogenous nitric oxide. J Biol Chem 2000, 275:28810-28815.

38 Horvath B, Banyasz T, Jian Z et al. Dynamics of the late Na(+) current during cardiac action potential and its contribution to afterdepolarizations. J Mol Cell Cardiol 2013, 64:59-68.

39 Wu L, Rajamani S, Li H et al. Reduction of repolarization reserve unmasks the proarrhythmic role of endogenous late Na(+) current in the heart. Am J Physiol Heart Circ Physiol 2009, 297:H1048-1057.

40 Wu L, Ma J, Li H et al. Late sodium current contributes to the reverse rate-dependent effect of IKr inhibition on ventricular repolarization. Circulation 2011, 123:1713-1720.

41 January CT, Riddle JM. Early afterdepolarizations: mechanism of induction and block. A role for L-type Ca2+ current. Circ Res 1989, 64:977-990.

42 Spencer CI, Sham JS. Effects of Na+/Ca2+ exchange induced by SR Ca2+ release on action potentials and afterdepolarizations in guinea pig ventricular myocytes. Am J Physiol Heart Circ Physiol 2003, 285:H2552-2562.

43 Antzelevitch C, Nesterenko V, Shryock JC et al. The role of late I Na in development of cardiac arrhythmias. Handb Exp Pharmacol 2014, 221:137-168.

44 Coppini R, Ferrantini C, Yao L et al. Late sodium current inhibition reverses electromechanical dysfunction in human hypertrophic cardiomyopathy. Circulation 2013, 127:575-584.

45 Makielski JC. Late sodium current: A mechanism for angina, heart failure, and arrhythmia. Trends Cardiovasc Med 2016, 26:115-122.

46 Flaim SN, Giles WR, McCulloch AD. Contributions of sustained INa and IKv43 to transmural heterogeneity of early repolarization and arrhythmogenesis in canine left ventricular myocytes. Am J Physiol Heart Circ Physiol 2006, 291:H2617-2629.

47 Zygmunt AC, Eddlestone GT, Thomas GP et al. Larger late sodium conductance in M cells contributes to electrical heterogeneity in canine ventricle. Am J Physiol Heart Circ Physiol 2001, 281:H689-697.

48 Antzelevitch C, Belardinelli L. The role of sodium channel current in modulating transmural dispersion of repolarization and arrhythmogenesis. J Cardiovasc Electrophysiol 2006, 17 Suppl 1:S79-S85.

49 Akalin F, Tirtir A, Yilmaz Y. Increased QT dispersion in epileptic children. Acta Paediatr 2003, 92:916-920.

50 Yamauchi M, Watanabe E, Yasui K et al. Prevention of ventricular extrasystole by mexiletine in patients with normal QT intervals is associated with a reduction of transmural dispersion of repolarization. Int J Cardiol 2005, 103:92-97.

51 Makielski JC, Farley AL. Na(+) current in human ventricle: implications for sodium loading and homeostasis. J Cardiovasc Electrophysiol 2006, 17 Suppl 1:S15-S20.

52 Wimmer NJ, Stone PH. Anti-anginal and anti-ischemic effects of late sodium current inhibition. Cardiovasc Drugs Ther 2013, 27:69-77.

53 Kumar K, Nearing BD, Bartoli CR et al. Effect of ranolazine on ventricular vulnerability and defibrillation threshold in the intact porcine heart. J Cardiovasc Electrophysiol 2008, 19:1073-1079.

54 Nieminen T, Nanbu DY, Datti IP et al. Antifibrillatory effect of ranolazine during severe coronary stenosis in the intact porcine model. Heart Rhythm 2011, 8:608-614.

55 Wu L, Guo D, Li H et al. Role of late sodium current in modulating the proarrhythmic and antiarrhythmic effects of quinidine. Heart Rhythm 2008, 5:1726-1734.

56 Wu L, Rajamani S, Shryock JC et al. Augmentation of late sodium current unmasks the proarrhythmic effects of amiodarone. Cardiovasc Res 2008, 77:481-488.

57 Wu L, Shryock JC, Song Y et al. An increase in late sodium current potentiates the proarrhythmic activities of low-risk QT-prolonging drugs in female rabbit hearts. J Pharmacol Exp Ther 2006, 316:718-726.

58 Antzelevitch C, Burashnikov A, Sicouri S et al. Electrophysiologic basis for the antiarrhythmic actions of ranolazine. Heart Rhythm 2011, 8:1281-1290.

59 Antzelevitch C, Belardinelli L, Wu L et al. Electrophysiologic properties and antiarrhythmic actions of a novel antianginal agent. J Cardiovasc Pharmacol Ther 2004, 9 Suppl 1:S65-83.

60 Antzelevitch C. Electrical heterogeneity, cardiac arrhythmias, and the sodium channel. Circ Res 2000, 87:964-965.

61 Burashnikov A, Antzelevitch C. Role of late sodium channel current block in the management of atrial fibrillation. Cardiovasc Drugs Ther 2013, 27:79-89.

62 Bacic D, Carneiro JS, Bento AA et al. Eleclazine, an Inhibitor of the Cardiac Late Sodium Current, Is Superior to Flecainide in Suppressing Catecholamine-Induced

Ventricular Tachycardia and T-Wave Alternans in an Intact Porcine Model. Heart Rhythm 2016.

63 Moss AJ, Zareba W, Schwarz KQ et al. Ranolazine shortens repolarization in patients with sustained inward sodium current due to type-3 long-QT syndrome. J Cardiovasc Electrophysiol 2008, 19:1289-1293.

64 Mazzanti A, Maragna R, Faragli A et al. Gene-Specific Therapy With Mexiletine Reduces Arrhythmic Events in Patients With Long QT Syndrome Type 3. J Am Coll Cardiol 2016, 67:1053-1058.

65 Scirica BM, Morrow DA, Hod H et al. Effect of ranolazine, an antianginal agent with novel electrophysiological properties, on the incidence of arrhythmias in patients with non ST-segment elevation acute coronary syndrome: results from the Metabolic Efficiency With Ranolazine for Less Ischemia in Non ST-Elevation Acute Coronary Syndrome Thrombolysis in Myocardial Infarction 36 (MERLIN-TIMI 36) randomized controlled trial. Circulation 2007, 116:1647-1652.

66 Liang F, Fan P, Jia J et al. Inhibitions of late INa and CaMKII act synergistically to prevent ATX-II-induced atrial fibrillation in isolated rat right atria. J Mol Cell Cardiol 2016, 94:122-130.

67 Carneiro JS, Bento AS, Bacic D et al. The Selective Cardiac Late Sodium Current Inhibitor GS-458967 Suppresses Autonomically Triggered Atrial Fibrillation in an Intact Porcine Model. J Cardiovasc Electrophysiol 2015, 26:1364-1369.

68 Fraser H, Belardinelli L, Wang L et al. Ranolazine decreases diastolic calcium accumulation caused by ATX-II or ischemia in rat hearts. J Mol Cell Cardiol 2006, 41:1031-1038.

69 Hale SL, Kloner RA. Ranolazine, an inhibitor of the late sodium channel current, reduces postischemic myocardial dysfunction in the rabbit. J Cardiovasc Pharmacol Ther 2006, 11:249-255.

70 Aldakkak M, Camara AK, Heisner JS et al. Ranolazine reduces Ca2+ overload and oxidative stress and improves mitochondrial integrity to protect against ischemia reperfusion injury in isolated hearts. Pharmacol Res 2011, 64:381-392.

71 Hale SL, Shryock JC, Belardinelli L et al. Late sodium current inhibition as a new cardioprotective approach. J Mol Cell Cardiol 2008, 44:954-967.

72 Zhang XQ, Yamada S, Barry WH. Ranolazine inhibits an oxidative stress-induced increase in myocyte sodium and calcium loading during simulated-demand ischemia. J Cardiovasc Pharmacol 2008, 51:443-449.

73 Chi L, Belardinelli L, Zeng A et al. Inhibition of late Na current, a novel target to improve diastolic function and electrical abnormalities in Dahl salt-sensitive rats. Am J Physiol Heart Circ Physiol 2016:ajpheart 00863 02015.

74 Rocchetti M, Sala L, Rizzetto R et al. Ranolazine prevents INaL enhancement and blunts myocardial remodelling in a model of pulmonary hypertension. Cardiovasc Res 2014, 104:37-48.

75 Rastogi S, Sharov VG, Mishra S et al. Ranolazine combined with enalapril or metoprolol prevents progressive LV dysfunction and remodeling in dogs with moderate heart failure. Am J Physiol Heart Circ Physiol 2008, 295:H2149-2155.

76 Chaitman BR, Skettino SL, Parker JO et al. Anti-ischemic effects and long-term survival during ranolazine monotherapy in patients with chronic severe angina. J Am Coll Cardiol 2004, 43:1375-1382.

77 Chaitman BR, Pepine CJ, Parker JO et al. Effects of ranolazine with atenolol, amlodipine, or diltiazem on exercise tolerance and angina frequency in patients with severe chronic angina: a randomized controlled trial. JAMA 2004, 291:309-316.

78 Stone PH, Gratsiansky NA, Blokhin A et al. Antianginal efficacy of ranolazine when added to treatment with amlodipine: the ERICA (Efficacy of Ranolazine in Chronic Angina) trial. J Am Coll Cardiol 2006, 48:566-575.

79 Kosiborod M, Arnold SV, Spertus JA et al. Evaluation of ranolazine in patients with type 2 diabetes mellitus and chronic stable angina: results from the TERISA randomized clinical trial (Type 2 Diabetes Evaluation of Ranolazine in Subjects With Chronic Stable Angina). J Am Coll Cardiol 2013, 61:2038-2045.

80 Morrow DA, Scirica BM, Karwatowska-Prokopczuk E et al. Effects of ranolazine on recurrent cardiovascular events in patients with non-ST-elevation acute coronary syndromes: the MERLIN-TIMI 36 randomized trial. JAMA 2007, 297:1775-1783.

81 Maier LS, Layug B, Karwatowska-Prokopczuk E et al. RAnoLazIne for the treatment of diastolic heart failure in patients with preserved ejection fraction: the RALI-DHF proof-of-concept study. JACC Heart Fail 2013, 1:115-122.

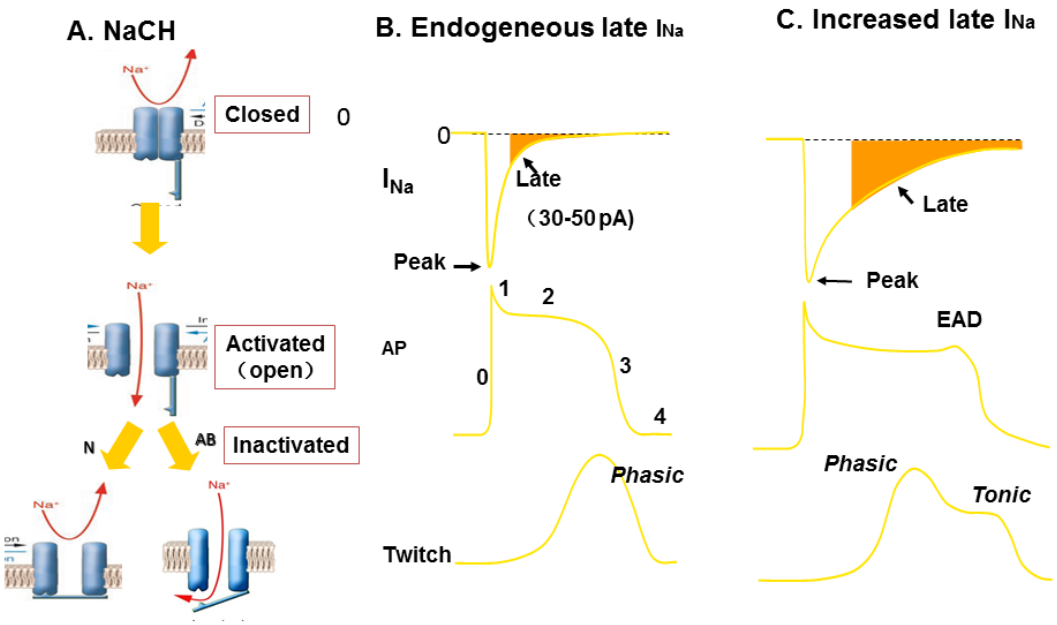
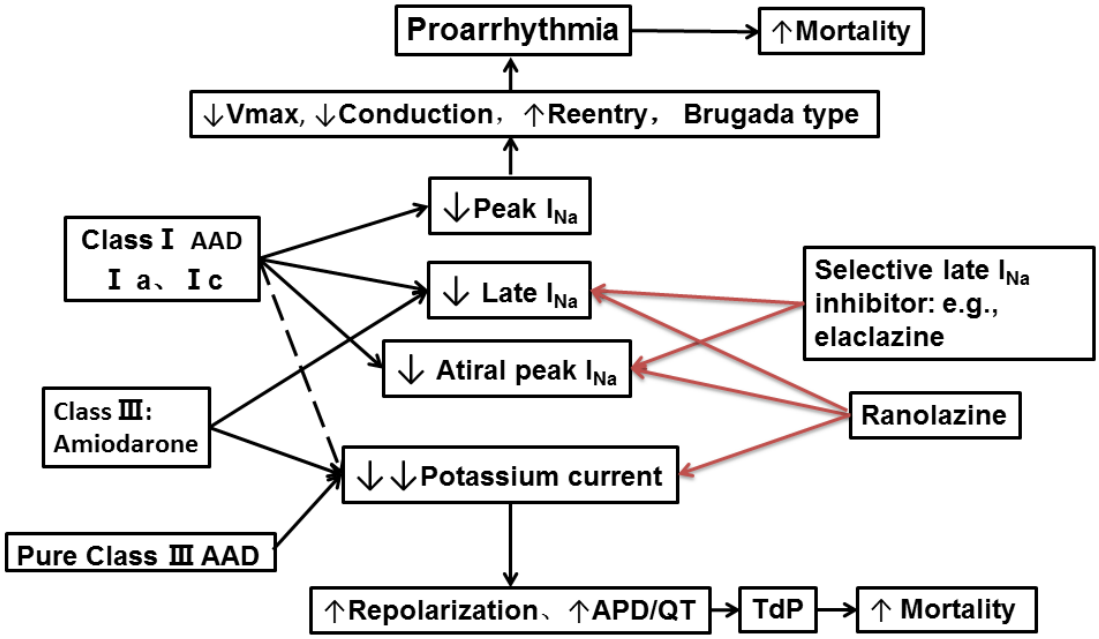


Figure 2


Proarrhythmia
↑Mortality
↓Vmax, ↓Conduction, ↑Reentry, Brugada type
↓Peak I_{Na}
Class Ⅰ AAD
Ⅰa、Ⅰc
↓ Late I_{Na}
Selective late I_{Na} inhibitor: e.g., elaclazine
↓ Atiral peak I_{Na}
Class Ⅲ: Amiodarone
Ranolazine
↓↓Potassium current
Pure Class Ⅲ AAD
↑Repolarization、↑APD/QT
TdP
↑ Mortality