Gender-based differences in cardiac diseases

Pei-Chi Yang*, Colleen E. Clancy
Department of Pharmacology, University of California Davis, Davis, CA 96516-5270, USA.
Received 26 October 2010, Revised 18 November 2010, Accepted 11 January 2011

Abstract

It has been observed that the incidence of heart failure and Brugada syndrome are higher in men, while women are more likely to have QT interval prolongation and develop torsades de pointes (TdP). Over the past decade, new studies have improved our understanding of the mechanisms of abnormal repolarization and the relationship between gender differences in cardiac repolarization and presentation of clinical syndromes. Nevertheless, the causes of gender-based differences in cardiac disease are still not completely clear. This review paper briefly summarized what is currently known about gender differences in heart failure, Brugada syndrome and long QT syndrome from molecular mechanisms to clinical presentations.

Keywords: gender differences, heart failure, Brugada syndrome, long QT

INTRODUCTION

In the past decade, it has become increasingly clear that cardiac arrhythmia and heart failure (HF) have gender-based differences that increase or reduce disease susceptibility[1-3]. The mechanisms of arrhythmia initiation, sustenance and termination as well as HF presentation appear to be gender specific. Recent clinical and experimental studies suggest that these differences may stem in part from fundamental intrinsic gender differences in cardiac tissue[4-10]. These include intrinsic electrical differences resulting from variable ion channel expression and diverse sex hormonal regulations via long-term genomic and acute non-genomic pathways[6,11-14], though the exact role gender plays in cardiac diseases is not fully understood.

GENDER-RELATED DIFFERENCES IN ELECTROPHYSIOLOGICAL REMODELING WITH HEART FAILURE

In HF, the heart cannot supply an adequate amount of blood to the rest of body. Blood moves to the heart and body at a slower rate, and pressure increases in the heart. In order to sustain cardiac performance, the chambers of the heart stretch to hold more blood to pump through the body by becoming thickened and stiff. For a short period of time, this helps to maintain the blood pressure, but eventually leads to cardiac dysfunction[15,16].

The common causes of HF include ischemic heart disease, cigarette smoking, hypertension, obesity, diabetes mellitus, and valvular heart disease. The causes of HF are difficult to analyze because of differences in gender, race and prevalence of causes changing with age. Clinical data confirm that HF is more common in patients older than 50 years[17] when testosterone levels are reduced. A number of studies have also found low levels of testosterone in HF patients[18], and have shown measurable short-term benefits from testosterone therapy[19,20]. However, no clear predictive role of testosterone levels has been defined. In addition, clinical trials have shown that the progression of HF is slower in women than in men, and females have im-
proved survival in HF\textsuperscript{[21-25]}. Compared to men, women tend to develop HF at older ages\textsuperscript{[26]}. Interestingly, women are more likely to develop diastolic HF with normal left ventricular ejection fraction compared with men\textsuperscript{[26-41]}

Thus, although sex differences have been observed in HF, the underlying mechanisms are still not clear. There are a number of recent detailed reviews on ion-channel remodeling in HF\textsuperscript{[27-30]} and gender differences in quality of life in HF patients\textsuperscript{[29,31-32]}. Here, we focus on gender differences in some of the major channels and transporters during electrophysiological remodeling.

It is well known that HF causes cardiac functional changes. These changes make the heart prone to arrhythmias and diastolic and systolic contractile dysfunction. One of the important regulators of cardiac contractile function is phospholamban (PLB). During systole, PLB binds to a Ca\textsuperscript{2+} pump and prevents Ca\textsuperscript{2+} from being pumped back into the sarcoplasmic reticulum (SR). During muscle relaxation, PLB is in its phosphorylated state, which removes its inhibitory effect on the SR Ca\textsuperscript{2+}-ATPase (SERCA) and restores low calcium levels in the cytoplasm\textsuperscript{[33]}. In a gene expression study, PLB is found highly expressed in human failing hearts\textsuperscript{[34]}, and may be a mechanism of systolic contractile dysfunction\textsuperscript{[35]}. Notably, in men, the expression levels of PLB are increased\textsuperscript{[36]}. PLB has also been shown to be phosphorylated by cAMP-dependent protein kinase and Ca\textsuperscript{2+}/calmodulin-dependent protein kinase\textsuperscript{[37,38]}. Calmodulin-3 has a lower expression level in men\textsuperscript{[39]}

The activity of the Na\textsuperscript{+}/K\textsuperscript{+}-ATPase via its interaction with the Na\textsuperscript{+}/Ca\textsuperscript{2+} exchanger (NCX) is important for maintaining Ca\textsuperscript{2+} homeostasis in the heart. HF studies have found reduced expression of Na\textsuperscript{+}/K\textsuperscript{+}-ATPase-\(\alpha_1\) in human failing heart tissue\textsuperscript{[34,42]}. This may lead to decreasing Ca\textsuperscript{2+} efflux by NCX, which increases cytoplasmic Ca\textsuperscript{2+} concentration and causes development of Ca\textsuperscript{2+}-dependent arrhythmias. In a gender difference study, it was discovered that men had reduced expression-levels of Na\textsuperscript{+}/K\textsuperscript{+}-ATPase-\(\alpha_1\)\textsuperscript{[36]}. In addition, the plasma membrane Ca\textsuperscript{2+}-ATPase isoform 4 was found to be less strongly expressed in HF mice\textsuperscript{[43]} and in men\textsuperscript{[30]}

Other cardiac functional changes in HF include action potential duration (APD) prolongation, reduction of cell excitability\textsuperscript{[44]}, increased Na\textsuperscript{+}/Ca\textsuperscript{2+} exchange, preserved \(\beta\)-adrenergic responsiveness, and reduced outward K\textsuperscript{+} currents (\(I_{\text{Ko}}\) or \(I_{\text{Ks}}\), which may contribute to APD prolongation\textsuperscript{[45]}. A HF study in porcine myocytes demonstrated that NCX is more phosphorylated in male pacing-induced failing swine and that \(\beta\)-adrenergic responsiveness was greatly reduced in males compared to females\textsuperscript{[46]}. This study suggested that increased NCX activity could lead to impaired contractile function by decreasing SR Ca\textsuperscript{2+} content and promote the development of arrhythmia triggers. Females may have better survival rates in HF because they have a smaller NCX current and larger preserved \(\beta\)-adrenergic regulation.

In ischemic myocytes, high levels of intracellular Na\textsuperscript{+} cause membrane potential changes that enhance Ca\textsuperscript{2+} influx via NCX. This increased influx could lead to Ca\textsuperscript{2+} “overload”. Various studies have been conducted to investigate the female gender in cardio-protection during ischemia and suggest a protective role of estrogen in hypertrophied and/or failing myocardium\textsuperscript{[47-51]}. One recent study showed that acute effects of estrogen at physiological concentration (1 nmol/L) reduced the increase in [Na\textsuperscript{+}], during metabolic inhibition (MI), and suggested that estrogen may regulate Ca\textsuperscript{2+} influx through reverse NCX by lessening the magnitude of the rise in [Na\textsuperscript{+}], during MI in ischemic hearts\textsuperscript{[47]}.

### BRUGADA SYNDROME AND MEN

East Asia is an area of high prevalence of Brugada syndrome (BrS), and the male-female ratio of the clinical phenotype is 8:1\textsuperscript{[52,53]}. BrS is a polymorphic ventricular tachycardia characterized by ST-segment elevation in the right precordial leads (V\textsubscript{1}-V\textsubscript{3}) and right bundle branch block. BrS has been linked to a number of mutations in the gene SCN5A encoding the cardiac Na\textsuperscript{+} channel, all of which cause loss of channel function\textsuperscript{[54-60]}

Two hypotheses have been discussed recently — the "repolarization hypothesis" and "depolarization hypothesis". The repolarization hypothesis is based on evidence for transmural dispersion of repolarization between the canine right ventricle (RV) epicardium and endocardium\textsuperscript{[61]}. Early repolarization due to loss of the AP-dome in the epicardium is expected to occur, which may induce phase 2-reentry and a substrate for the development of VT/VF\textsuperscript{[50,62-63]}

On the other hand, the depolarization hypothesis proposed by Wild and Postema suggests RV conduction delay as part of the pathophysiologic mechanism of BrS that is supported by clinical data\textsuperscript{[64]}. They propose that depolarization abnormalities with mild structural abnormalities\textsuperscript{[65]} may explain the clinical observed repolarization abnormalities on the electrocardiogram (ECG). Antzelevitch et al\textsuperscript{[60]}. agree that slowed conduction and mild structural defects exist in some BrS cases, especially in \(I_{\text{Ko}}\) loss of function cases, but it is not absolutely required. They pointed out that noticeable accentuation of the epicardial AP notch can
account for the ST segment elevation associated with BrS by causing loss of the AP dome in some cells and not others, leading to a dispersion of repolarization. Dispersion of repolarization might allow a premature beat to trigger reentrant arrhythmias.

An ionic and cellular basis for the intriguing sex-related distinction in presentation of BrS was first proposed by Di Diego and colleagues. They suggested that a more prominent $I_{\text{ka}}$ in males leads to the predominance of Brugada phenotype in men. In a recent ion-channel expression-pattern study, Gaborit et al. found lower-level expression of repolarizing ion-channel subunits including KCNIP2, HERG, Kir2.3, Kir6.2 and SUR2 in females that may protect them against the Brugada phenotype (but also make them susceptible to long QT as discussed below). In male RV-epicardium, higher-level expression of the $I_{\text{ka}}$ β-subunit KCNIP2 has been observed. Larger male current may favor early repolarization in Brugada-patients. However, no gender-differences were found in epicardial KCNIP2-expression in human LV (Fig. 1). This finding is compatible with a canine model of sex-related differences conducted by Di Diego et al., showing no sex differences on $I_{\text{ka}}$ current and epicardial phase-1 repolarization in LV.

LONG QT SYNDROME AND WOMEN

Female gender is a determinant of susceptibility to certain types of cardiac arrhythmia. For example, female sex is a risk factor for inherited and acquired long-QT (LQT) syndrome and associated with torsade de pointes (TdP) arrhythmias. Various studies have shown that females have a higher risk of a first cardiac event between 15 and 40 years, and observed that women are at higher risk than men of drug-induced TdP by class III anti-arrhythmic drugs and other drugs that block HERG. Animal studies have shown higher-level inward currents in females. These agree with a recent expression-pattern study, where the authors found lower expression-levels of $I_{\text{Ks}}$ channel α- (Kir2.3, Kv1.4 and HERG) and β- (minK) subunits in female heart. The differences between male and female HERG were significant in RV only, while the sex differences on Kir2.3, Kv1.4 and minK were significant in both RV and LV (Fig. 2).

The fact that women are at particular risk for drug-induced arrhythmias and that arrhythmia risk rises around the time of puberty, suggests the dominant female hormones estrogen and progesterone modulate arrhythmia vulnerability. While estrogen may exacerbate arrhythmia susceptibility by directly interacting with the drug binding site on the promiscuous hERG subunit and reducing $I_{\text{Ka}}$ current and increasing the rate of channel deactivation, progesterone is apparently protective and reduces QT intervals. Studies suggested that both progesterone and testosterone acutely modulate $I_{\text{Ka}}$ and $I_{\text{CaL}}$ through phosphoinositide 3-kinase (PI3K)/AKT-dependent endothelial nitric oxide (NO) synthase (eNOS) activation pathway.
resulting in suppressing $I_{CaL}$ currents and increasing $I_{Ks}$ current density.

It has been recently suggested that the N-terminal truncated isoform of the androgen receptor (AR45) plays an essential role in the heart since the transcript level of the AR45 is high in human heart tissue. An experiment of AR45 effects on the HERG potassium channel demonstrated that AR45 enhanced HERG channels by stabilizing HERG channel protein via ERK1/2 stimulations\[83\]. Other studies also indicated that the male hormone testosterone (5a-DHT) increased repolarizing $K^+$ currents density ($I_{K1}$ and $I_{Kr}$) and acts to protect against arrhythmia initiation\[11,74,78,84\].

Nakagawa et al.\[85\] have observed that during the follicular phase (prior to ovulation) of the menstrual cycle, QT interval is longer than that in the luteal phase (following ovulation) when progesterone is increased. Arrhythmic events associated with acquired and inherited LQTs are significantly reduced during phases where progesterone level is high\[85\]. Moreover, QT is significantly increased by estrogen hormone replacement therapy in females and susceptibility to drug-induced arrhythmias is exaggerated in the late follicular phase where estrogen level is the highest\[85\]. In contrast, Burke et al.\[86\] found that in pre-menopausal women the corrected QT (QTc) interval does not greatly change through the menstrual cycle, but QTc is reduced in the luteal phase after autonomic blockade. Furthermore, one study showed that QTc did not change during the menstrual cycle, but its shortening was more pronounced in the luteal phase with ibutilide application in women\[87\]. The disparity in these studies may be due to the fact that corrected QT interval measurements were based on a single point or a few points with the individual patient at rest. Such an analysis is unlikely to be sensitive enough to observe

**Fig. 2 Expression-profile of gender-differential $K^+$-channel genes.** The same format as in Fig. 1. The results are expressed as mean±SD from 7 donors/gender. In both the EPI and ENDO, the inward-rectifier Kir6.2 and Kir2.3 were weakly expressed in women. The regulator of Kir6.2, SUR2, was lower in female right ventricle (RV). The rapid delayed-rectifier HERG was strongly expressed in male hearts in both the EPI and ENDO. In female, the $I_{Ks}\beta$-subunit minK was expressed at lower levels, and Kv1.4 was strongly expressed in male ENDO. $P < 0.05$, $***P < 0.001$ vs EPI; a: $P < 0.05$, b: $P < 0.01$ vs women. LV-statistics: EPI vs ENDO: male, Kv1.4, $P < 0.01$; female, Kv1.4, $P < 0.05$, Kir2.3, $P < 0.01$. Male EPI vs Female: Kir2.3, $P < 0.001$; HERG, $P < 0.05$; minK, $P < 0.01$; Kv1.4, $P < 0.01$. Male ENDO vs Female: Kir2.3, $P < 0.05$; minK, $P < 0.01$; Kv1.4, $P < 0.05$. EPI: epicardium; ENDO: endocardium. Reprinted with permission from Gaborit et al. (2010)\[36\] with modification.
significant individual differences in QT intervals as they fluctuate throughout the menstrual cycle since biological variability between patients may be larger than fluctuations in individual patients.

In addition, some drug studies demonstrated that females have greatly increased QT intervals compared with males during treatment with d, l-sotalol \[^{[72]}\] and quinidine \[^{[88-90]}\]. \(I_{Kr}\) blockers seem to increase early after depolarization (EAD) development and prolong repolarization in females, both primary and critical predictors of drug-induced TdP \[^{[91,92]}\].

**SIMULATION APPROACH TO UNDERSTAND EFFECTS OF SEX STEROID HORMONES AND DRUGS**

It is challenging to determine the role of gender experimentally in complex cardiac functioning since gender effects are multi-factorial and affect cardiac components at different scales of the cardiac system. However, a computational approach can be useful in this respect as it allows study of specific effects in isolation without other perturbations to the system. For example, it is not easy to determine how much a role physiological concentrations of circulating sex steroid hormones play in gender linked arrhythmia susceptibility. Computational models can incorporate the effects of sex hormones measured experimentally and test these changes specifically from non-linear interactions within cells, between cells and among various tissue components that culminate to produce the overall effects of gender on the heart. In this case, simulations can be used to investigate how acute sex hormones and drugs affect system behavior \[^{[93]}\]. The tissue simulations shown in [Fig. 3](#) predict the effects of sex steroid hormones on clinically observed QT intervals and on drug-induced LQTS. Estrogen significantly increases susceptibility to drug-induced arrhythmias. However, low concentrations of testosterone are sufficient to protect against drug-induced arrhythmias (Fig. 3). Our simulation studies have resulted in improved understanding of mechanisms of estrogen-mediated susceptibility to drug-induced arrhythmia initiation \[^{[93]}\] and protective effects of progesterone and testosterone against congenital and drug-induced LQT syndrome \[^{[82,93]}\]. Moreover, theoretical studies have revealed gender effects at the cellular and tissue-levels as well as predicted effects of sex steroid hormone on the body surface by computing "pseudo" electrocardiograms \[^{[82,93]}\].
Fig. 3 2D heterogeneous tissue simulations during short-long-short pacing protocols. Four snapshots following application of hormones and/or drug at indicated time points. Tissues were stimulated along one edge and propagated from the endocardial to the epicardial region followed by a point stimulus applied in the right edge of the endocardial region. Voltages are indicated by color gradient. A: In the absence of hormones or drugs, no reentry occurs (first row). The same behavior is observed following drug application alone (E-4031) and with testosterone application alone (DHT 10 nmol/L). However, when estrogen (E2) (1 nmol/L) is present (bottom row), the reentry was induced. B: Comparison of 2D heterogeneous tissue dynamics in the absence or presence of E-4031 during the late follicular phase, and application of testosterone 3 nmol/L with E-4031 addition. The simulation results show no reentrant activity during the late follicular phase of the menstrual cycle (progesterone, 2.5 nmol/L and E2, 1 nmol/L). However, during the late follicular phase, a spiral wave is readily induced when 10 nmol/L E-4031 is applied. Finally, testosterone 3 nmol/L with 10 nmol/L E-4031 did not trigger reentry activity. C: The same protocol as above was used, but the premature stimulus was applied during the vulnerable window in the middle of the endocardial tissue near the boundary between the endocardial region and M cells. The late follicular phase with E-4031 is shown. Reentry was introduced in this condition. The results show the effect of a point stimulus applied in the middle of the endocardial tissue, leading to the initiation of a pair of counter-rotating spiral waves. EPI: picardium; ENDO: endocardium. Reproduced with permission from Yang et al. (2010) with modification.

References

[1] Andelfinger G, Tapper AR, Welch RC, Vanoye CG, George AL Jr, Benson DW. KCNJ2 mutation results in Andersen syndrome with sex-specific cardiac and skeletal muscle phenotypes. Am J Hum Genet 2002;71:663-8.

[2] Arya A. Gender-related differences in ventricular repolarization: beyond gonadal steroids. J Cardiovasc Electrophysiol 2005;16:525-7.

[3] Teplitz L, Igic R, Berbaum ML, Schwertz DW. Sex differences in susceptibility to epinephrine-induced arrhythmias. J Cardiovasc Pharmacol 2005;46:548-55.

[4] Di Diego JM, Cordeiro JM, Goodrow RJ, Fish JM, Zymunt AC, Perez GI, et al. Ionic and cellular basis for the predominance of the Brugada syndrome phenotype in males. Circulation 2002;106:2004-11.

[5] Fish JM, Antzlelevitch C. Cellular and ionic basis for the sex-related difference in the manifestation of the Brugada syndrome and progressive conduction disease phenotypes. J Electrocardiol 2003;36(Suppl):S173-9.

[6] Xiao L, Zhang L, Han W, Wang Z, Nattel S. Sex-based transmural differences in cardiac repolarization and
ion-current properties in canine left ventricles. Am J Physiol Heart Circ Physiol 2006;291:H570-80.

[7] Pham TV, Robinson RB, Danilo P, Jr., Rosen MR. Effects of gonadal steroids on gender-related differences in transmural dispersion of L-type calcium current. Cardiovasc Res 2002;53:752-62.

[8] Hara M, Danilo P, Jr., Rosen MR. Effects of gonadal steroids on ventricular repolarization and response on the right E1031. J Pharmacol Exp Ther 1998;285:1069-72.

[9] Pham TV, Rosen MR. Sex, hormones, and repolarization. Cardiovasc Res 2002;53:740-51.

[10] Gowda RM, Khan LA, Pumukkolla G, Vasavada BC, Sacchi TJ, Wilbur SL. Female preponderance in ibutilide-induced torsade de pointes. Int J Cardiol 2004;95:219-22.

[11] Bai CX, Kurokawa J, Tamagawa M, Nakaya H, Furukawa T. Nontranscriptional regulation of cardiac repolarization currents by testosterone. Circulation 2005;112:1701-10.

[12] Furukawa T, Kurokawa J. Regulation of cardiac ion channels via non-genomic action of sex steroid hormones: implication for the gender difference in cardiac arrhythmias. Pharmacol Ther 2007;115:106-15.

[13] Korte T, Fuchs M, Arkudas A, Geertz S, Meyer R, Gardiwal A, et al. Female mice lacking estrogen receptor beta display prolonged ventricular repolarization and reduced ventricular automaticity after myocardial infarction. Circulation 2005;111:2282-90.

[14] Nakagawa M, Ooie T, Ou B, Ichinose M, Takahashi N, Hara M, et al. Gender differences in autonomic modulation of ventricular repolarization in humans. J Cardiovasc Electrophysiology 2005;16:278-84.

[15] What is heart failure? [cited 2010 Sep. 20]; Available from: http://www.americanheart.org/print_presenter.jhtml?identifier=337

[16] Definition of Heart failure. June 18, 2002 [cited 2010 Sep. 20]; Available from: http://www.medterms.com/script/main/art.asp?articlekey=3672

[17] Heart Disease and Stroke Statistics -- 2010 Update. [cited 2010 Oct. 22]; Available from: http://americanheart.org/presenter.jhtml?identifier=3000090

[18] Theodoraki A, Bouloux PM. Low sex hormones in heart failure. Heart 2010;96:946-7.

[19] Malkin CJ, Pugh PJ, West JN, van Beek EJ, Jones TH, Channer KS. Testosterone therapy in men with moderate severity heart failure: a double-blind randomized placebo controlled trial. Eur Heart J 2006;27:57-64.

[20] Caminiti G, Volterrani M, Iellamo F, Marazzi G, Masaro R, Miceli M, et al. Effect of long-acting testosterone treatment on functional exercise capacity, skeletal muscle performance, insulin resistance, and baroreflex sensitivity in elderly patients with chronic heart failure a double-blind, placebo-controlled, randomized study. J Am Coll Cardiol 2009;54:919-27.

[21] Adams KF, Jr., Sueta CA, Gheorghiade M, O’Connor CM, Schwartz TA, Koch GG, et al. Gender differences in survival in advanced heart failure. Insights from the FIRST study. Circulation 1999;99:1816-21.

[22] Ho KK, Anderson KM, Kannel WB, Grossman W, Levy D. Survival after the onset of congestive heart failure in Framingham Heart Study subjects. Circulation 1993;88: 107-15.

[23] Adams KF Jr, Dunlap SH, Sueta CA, Clarke SW, Patterson JH, Blauwert MB, et al. Relation between gender, etiology and survival in patients with symptomatic heart failure. J Am Coll Cardiol 1996;28:1781-8.

[24] Croft JB, Giles WH, Pollard RA, Keenan NL, Casper ML, Anda RF. Heart failure survival among older adults in the United States: a poor prognosis for an emerging epidemic in the Medicare population. Arch Intern Med 1999;159:505-10.

[25] Simon T, Mary-Krause M, Funck-Brentano C, Jaillon P. Sex differences in the prognosis of congestive heart failure: results from the Cardiac Insufficiency Bisoprolol Study (CIBIS II). Circulation 2001;103:375-80.

[26] Stromberg A, Martensson J. Gender differences in patients with heart failure. Eur J Cardiovasc Nurs 2003;2:7-18.

[27] Wang Y, Hill JA. Electrophysiological remodeling in heart failure. J Mol Cell Cardiol 2010;48:619-32.

[28] Tomasselli GF, Zipes DP. What causes sudden death in heart failure? Circ Res 2004;95:754-63.

[29] Nattel S, Maguy A, Le Bouter S, Yeh YH. Arrhythmogenic ion-channel remodeling in the heart: heart failure, myocardial infarction, and atrial fibrillation. Physiol Rev 2007;87:425-56.

[30] Noss RD, Aiba T, Tomasselli GF, Akar FG. Mechanisms of disease: ion channel remodeling in the failing ventricle. Nat Clin Pract Cardiovasc Med 2008;5:196-207.

[31] Heo S, Moser DK, Widener J. Gender differences in the effects of physical and emotional symptoms on health-related quality of life in patients with heart failure. Eur J Cardiovasc Nurs 2007;6:146-52.

[32] Riedinger MS, Dracup KA, Brecht ML, Padilla G, Sarna L, Ganz PA. Quality of life in patients with heart failure: do gender differences exist? Heart Lang 2001;30:105-16.

[33] Frank K, Kranias EG. Phospholamban and cardiac contractility. Ann Med 2000;32:572-8.

[34] Borlak J, Thum T. Hallmarks of ion channel gene expression in end-stage heart failure. FASEB J 2003;17:1592-608.

[35] Freeman K, Lerman I, Kranias EG, Bohlmeyer T, Bristow MR, Lefkowitz RJ, et al. Alterations in cardiac adrenergic signaling and calcium cycling differentially affect the progression of cardiomyopathy. J Clin Invest 2001;107:967-74.

[36] Gaborit N, Varro A, Le Bouter S, Szuts V, Escande D, Nattel S, et al. Gender-related differences in ion-channel and transporter subunit expression in non-diseased human hearts. J Mol Cell Cardiol 2010;49:639-46.

[37] Yang D, Song LS, Zhu WZ, Chakir K, Wang W, Wu C, et al. Calmodulin regulation of excitation-contraction coupling in cardiac myocytes. Circ Res 2003;92:659-67.
[38] Lindemann JP, Watanabe AM. Phosphorylation of phospholamban in intact myocardium. Role of Ca2+-calmodulin-dependent mechanisms. *J Biol Chem* 1985;260: 4516-25.

[39] Redfield MM, Jacobsen SJ, Burnett JC, Jr., Mahoney DW, Bailey KR, Rodeheffer RJ. Burden of systolic and diastolic ventricular dysfunction in the community: appreciating the scope of the heart failure epidemic. *JAMA* 2003;289:194-202.

[40] Vasan RS, Larson MG, Benjamin EJ, Evans JC, Reiss CK, Levy D. Congestive heart failure in subjects with normal versus reduced left ventricular ejection fraction: prevalence and mortality in a population-based cohort. *J Am Coll Cardiol* 1999;33:1948-55.

[41] Masoudi FA, Havranek EP, Smith G, Fish RH, Steiner DF, et al. Gender, age, and heart failure with preserved left ventricular systolic function. *J Am Coll Cardiol* 2003;41:217-23.

[42] Schwinger RH, Wang J, Frank K, Muller-Ehmsen J, Brixius K, McDonough AA, et al. Reduced sodium pump alpha1, alpha3, and beta1 isoform protein levels and Na+/K+ ATPase activity but unchanged Na+/Ca2+ exchanger protein levels in human heart failure. *Circulation* 1999;99:2105-12.

[43] Wu X, Chang B, Blair NS, Sargent M, York AJ, Robbins J, et al. Plasma membrane Ca2+-ATPase isoform 4 antagonizes cardiac hypertrophy in association with calcineurin inhibition in rodents. *J Clin Invest* 2009;119:976-85.

[44] Vermeulen JT. Mechanisms of arrhythmias in heart failure. *J Cardiovasc Electrophysiol* 1998;9:208-21.

[45] Pogwizd SM, Bers DM. Cellular basis of triggered arrhythmias in heart failure. *Trends Cardiovasc Med* 2004;14:61-6.

[46] Wei SK, McCurley JM, Hanlon SU, Haigney MC. Gender differences in Na/Ca exchanger current and beta-adrenergic responsiveness in heart failure in pig myocytes. *Ann N Y Acad Sci* 2007;1099:183-9.

[47] Sugishita K, Su Z, Li F, Philipson KD, Barry WH. Gender influences [Ca(2+)](i) during metabolic inhibition in myocytes overexpressing the Na(+)-Ca(2+) exchanger. *Circulation* 2001;104:2101-6.

[48] Cross HR, Lu L, Steenbergen C, Philipson KD, Murphy E. Overexpression of the cardiac Na(+)/Ca(2+) exchanger increases susceptibility to ischemia/reperfusion injury in male, but not female, transgenic mice. *Circ Res* 1998;83:1215-23.

[49] Jovanovic S, Jovanovic A, Shen WK, Terzic A. Low concentrations of 17beta-estradiol protect single cardiac cells against metabolic stress-induced Ca2+ loading. *J Am Coll Cardiol* 2000;36:948-52.

[50] Fraser H, Davidge ST, Clanachan AS. Enhancement of post-ischemic myocardial function by chronic 17 beta-estradiol treatment: role of alterations in glucose metabolism. *J Mol Cell Cardiol* 1999;31:1539-49.

[51] Keller JN, Germeyer A, Begley JG, Mattson MP. 17Beta-estradiol attenuates oxidative impairment of synaptic Na+/K+-ATPase activity, glucose transport, and glutamate transport induced by amyloid beta-peptide and iron. *J Neurosci Res* 1997;50:522-30.

[52] Sarkozy A, Brugada P. Sudden cardiac death and inherited arrhythmia syndromes. *J Cardiovasc Electrophysiol* 2005;16 (Suppl 1):S8-20.

[53] Wilde AA, Antzelevitch C, Borggrefe M, Brugada J, Brugada R, Brugada P, et al. Proposed diagnostic criteria for the Brugada syndrome: consensus report. *Circulation* 2002;106:2514-9.

[54] Clancy CE, Rudy Y. Na(+) channel mutation that causes both Brugada and long-QT syndrome phenotypes: a simulation study of mechanism. *Circulation* 2002;105:1208-13.

[55] Grant AO, Carboni MP, Neplioueva V, Starmer CF, Memmi M, Napolitano C, et al. Long QT syndrome, Brugada syndrome, and conduction system disease are linked to a single sodium channel mutation. *J Clin Invest* 2002;110:1201-9.

[56] Benson DW, Wang DW, Dymt M, Knilans TK, Fish FA, Strieper MJ, et al. Congenital sick sinus syndrome caused by recessive mutations in the cardiac sodium channel gene (SCN5A). *J Clin Invest* 2003;112:1019-28.

[57] Bennett PB, Yazawa K, Makita N, George AL, Jr. Molecular mechanism for an inherited cardiac arrhythmia. *Nature* 1995;376:683-5.

[58] Tan HL, Bink-Boelkens MT, Bezzina CR, Viswanathan PC, Beaufort-Krol GC, van Tintelen PJ, et al. A sodium-channel mutation causes isolated cardiac conduction disease. *Nature* 2001;409:1043-7.

[59] Veldkamp MW, Viswanathan PC, Bezzina C, Baartscheer A, Wilde AA, Balser JR. Two distinct congenital arrhythmias evoked by a multidysfunctional Na(+) channel. *Circ Res* 2000;86:E91-7.

[60] Chen Q, Kirsch GE, Zhang D, Brugada R, Brugada J, Brugada P, et al. Genetic basis and molecular mechanism for idiopathic ventricular fibrillation. *Nature* 1998;392:293-6.

[61] Yan GX, Antzelevitch C. Cellular basis for the Brugada syndrome and other mechanisms of arrhythmogenesis associated with ST-segment elevation. *Circulation* 1999;100:1660-6.

[62] Antzelevitch C, Brugada P, Brugada J, Brugada R, Shimizu W, Gussak I, et al. Brugada syndrome: a decade of progress. *Circ Res* 2002;91:1114-8.

[63] Antzelevitch C, Brugada P, Brugada J, Towbin JA, Nademane K. Brugada syndrome: 1992-2002: a historical perspective. *J Am Coll Cardiol* 2003;41:1665-71.

[64] Wilde AA, Postema PG, Di Diego JM, Viskin S, Morita H, Fish JM, et al. The pathophysiological mechanism underlying Brugada syndrome: depolarization versus repolarization. *J Mol Cell Cardiol* 2010;49:543-53.

[65] Meregalli PG, Wilde AA, Tan HL. Pathophysiological mechanisms of Brugada syndrome: depolarization disorder, repolarization disorder, or more? *Cardiovasc Res* 2005;67:367-78.
