In the 1880’s, Sydney Ringer confirmed that calcium (Ca\(^{2+}\)) was an essential second messenger for the heart. It is also a key link between data collected from laboratories (bench) and therapeutic application (bedside) for patients with diseased hearts. Abnormal Ca\(^{2+}\) handling by the sarcoplasmic reticulum (SR), the storage organelle in cardiomyocytes, has been shown to play a crucial role in the pathogenesis of heart failure and lethal arrhythmia. Mutation within the 2 major Ca\(^{2+}\)-regulatory proteins in the SR [ie, the cardiac ryanodine receptor (RyR2) (Figure) and calsequestrin (CASQ2)] is known to cause aberrant Ca\(^{2+}\) release during diastole via RyR2, upon β-adrenergic stimulation (ie, induced by exercise or emotional stress); this phenomenon is known as “diastolic Ca\(^{2+}\) leak”. Ca\(^{2+}\) leak provides a substrate for delayed afterdepolarization.

**New Data on Catecholaminergic Polymorphic Ventricular Tachycardia in Japan**

– From the Bench to the Bedside –

Shinichi Okuda, MD, PhD; Masafumi Yano, MD, PhD

---

The opinions expressed in this article are not necessarily those of the editors or of the Japanese Circulation Society.

Received May 14, 2013; accepted May 15, 2013; released online May 25, 2013

Department of Medicine and Clinical Science, Division of Cardiology, Yamaguchi University Graduate School of Medicine, Ube, Japan

Mailing address: Shinichi Okuda, MD, PhD, Department of Medicine and Clinical Science, Division of Cardiology, Yamaguchi University Graduate School of Medicine, 1-1-1 Minamikogushi, Ube 755-8505, Japan. E-mail: sokuda@yamaguchi-u.ac.jp

ISSN-1346-9843 doi: 10.1253/circj.CJ-13-0623

All rights are reserved to the Japanese Circulation Society. For permissions, please e-mail: cj@j-circ.or.jp
which leads to syncope and life-threatening arrhythmia at a younger age; these conditions together comprise catecholaminergic polymorphic ventricular tachycardia (CPVT).1

Genetic Characteristics and Pathologic Mechanisms of CPVT
Since 15 years ago, when a point mutation within RyR2 was first reported to be linked to CPVT (CPVT1), more than 150 RyR2 missense mutations have been identified, mostly in studies from Europe and the United States.2-6 More than 70% of the mutations cluster in 3 regions of the RyR2 channel, which are designated as the N-terminal domain, central domain, and C-terminal domain.1 Mutations at different positions in either domain seem to result in a gain of function; that is, hyperactivation of RyR2 and hypersensitization against agonists. In this issue of the Journal, Kawamura et al7 provide valuable genetic and clinical data from 50 Japanese probands with CPVT, including the discovery of new mutation sites. In agreement with previous reports from Western countries,2-6 almost all probands were of a younger age (<15 years), and almost all missense mutations of RyR2 clustered in the 3 abovementioned restricted domains in their patients with CPVT. Approximately 46% of mutations noted in the Japanese probands were found near the C-terminal region. Interestingly, the distribution rate of the mutation within the N-terminal domain of RyR2 (approximately 11%) appears to be lower than that reported in previous studies (approximately 20%),2,4 although there was no statistically significant difference. The reason for this discrepancy is not clear, but it could be partially attributed to the small number of patients in the study or to racial differences. The reason for this discrepancy is not clear, but it could be partially attributed to the small number of patients in the study or to racial differences. The reason for this discrepancy is not clear, but it could be partially attributed to the small number of patients in the study or to racial differences. The reason for this discrepancy is not clear, but it could be partially attributed to the small number of patients in the study or to racial differences.

Three hypotheses have been proposed to explain dysregulation of RyR2 channel gating in CPVT.1
(1) Defective interdomain interaction between the N-terminal and central domain of RyR2. In a normal channel, the N-terminal and central domain interact with each other to act as a regulatory switch for channel-gating activity. A mutation in either domain weakens the interdomain interaction, thus increasing the tendency towards unzipping, which causes activation and leakiness of RyR2.1
(2) Dissociation of FKBP binding protein 12.6 (FKBP12.6) from RyR2.4 In a normal channel, FKBP12.6 provides stability by binding RyR2 tightly. In the failing heart, hyperphosphorylation of RyR2 by protein kinase A occurs during excessive β-adrenergic stimulation, which causes dephosphorylation of FKBP12.6 from RyR2 and consequently destabilization of RyR2. RyR2 mutations have been associated with altered FKBP12.6 binding..
(3) Store overload-induced Ca2+ release (SOICR).2 The threshold of SR Ca2+ load for channel activation (determined by Ca2+ sparks), regulated by luminal [Ca2+]i, not cytosolic [Ca2+]i in the SR, is markedly reduced in mutated RyR2 and CASQ2.2 New findings obtained from the partial crystal structure of the N-terminal domain and central domain (including the phosphorylation site) of RyR2 might be helpful for gaining further insights on this topic.7,8

A point mutation within CASQ2 is another cause of CPVT (CPVT2). Thus far, 14 CASQ2 mutations of CPVT have been described.2 Mutations in CASQ2 may induce defective protein-protein interactions in a quaternary complex of RyR2, CASQ2, triadin, and junction, which results in activation of RyR2 function, sensitized by luminal [Ca2+]i activation during β-adrenergic stimulation.1,2 Kawamura et al7 report that 1 proband with CPVT2, who had very severe symptoms, was a compound heterozygous mutation carrier of CASQ2, thus suggesting the importance of mutations within CASQ2 for development of the severe phenotype. This is the second case of a compound heterozygous mutation carrier of CASQ2. New gene mutations within the Kir2.1 channel were identified as CPVT3, and another syndrome related to CPVT and long QT (ie, Ankyrin-B disease) were also identified, although the underlying mechanism remains to be determined.

Therapeutic Approach to and Future Measures Against CPVT
The therapeutic approach to CPVT is rooted in improving the abnormal Ca2+ handling during β-adrenergic stimulation.1,2,6 Beta-blocker therapy reduces the heart rate and β-adrenergic tone, as well as increasing the possibility of improved Ca2+ handling in diseased hearts.9 In agreement with previous reports,3,4 the study by Kawamura et al7 showed that more than 90% of patients with CPVT in Japan were treated with β-blockers, but in 19% of them it was impossible to prevent syncope or lethal arrhythmias under this treatment. In their study, the mean observation period was approximately 4 years. A longer-term survival rate after appropriate therapeutic strategies should be determined. Although implantable cardioverter defibrillators (ICD) are known to prevent sudden cardiac death caused by life-threatening arrhythmia, there are some limitations to the effective use of ICD for CPVT patients, owing to the increase in β-adrenergic tone by ICD shocks and the technical complication of ICD implantation in young patients. For patients who are resistant to β-blocker therapy, flecainide and/or Ca2+ channel antagonists seem to be partly effective as adjunctive therapy.10 Flecainide, a Class IC antiarrhythmic drug with Na+ channel-blocking effect, in combination with β-blockers, can reduce the incidence of exercise-induced ventricular arrhythmias in CPVT patients.11 The mechanism for the prevention of CPVT by flecainide is still unclear. A direct action of flecainide against RyR2 in order to prevent aberrant Ca2+ release has been recently proposed.12 However, it has been also shown that flecainide prevents triggered activity by blocking Na+ channels without directly affecting RyR2.13 To reduce β-adrenergic tone, left cardiac sympathetic denervation has been used clinically in Europe.14 An “RyR2 stabilizer” may be a new candidate for future strategies for the prevention of CPVT. Both dantrolene,15 which specifically binds to the N-terminal domain of RyR2, and K201 (JTV519),1 which binds to the central domain of RyR2, inhibit Ca2+ leak by correcting the defective interdomain interaction between the N-terminal and central domain of RyR2.16 S107, a K201 derivative, has been reported to increase the binding affinity of FKBP12.6 for RyR2, resulting in prevention of CPVT in a knock-in (KI) mouse model with a mutation in the central domain of RyR2 (R2474S+/–).17 VK-II-86, a carvedilol analog with lesser β-blockade efficiency than carvedilol, has been reported to shorten the duration of inducible VT after epinephrine plus caffeine injection in a RyR2-R4496C+/– KI mouse model, by preventing SOICR.18 Propafenone,17 a Class IC antiarrhythmic drug with a β-blocker-like effect, prevented ventricular arrhythmia in a CASQ2-deficient CPVT mouse model.

Basic and clinical research is currently in progress in this area. Ca2+ directly regulates arrhythmogenesis in diseased hearts, resulting in lethal arrhythmia. Because β-blocker therapy has several limitations, development of other therapies for CPVT should be prioritized. Additionally, the knowledge gained from various studies of mutated RyR2 and CASQ2 under

Article p1705

New Data on CPVT in Japan
pathological conditions should be applied to the identification of potentially useful therapeutic agents that inhibit diastolic Ca\textsuperscript{2+} leak. Thus, Ca\textsuperscript{2+} could be a key factor linking the basic data and clinical manifestations of CPVT and aid in the development of a new strategy against CPVT. Progression from molecular discoveries to the use of those molecules as therapeutic agents is currently underway, but the end goal of clinical application is yet to be achieved. Future studies should focus on determining the most efficient therapy for improving the survival rate and reducing the rate of severe life-threatening symptoms.

References

1. Yano M, Yamamoto T, Ikeda Y, Matsuzaki M. Mechanisms of disease: Ryanodine receptor defects in heart failure and fatal arrhythmia. Nat Clin Pract Cardiovasc Med 2006; 3: 43–52.

2. Priori SG, Chen SR. Inherited dysfunction of sarcoplasmic reticulum Ca\textsuperscript{2+} handling and arrhythmogenesis. Circ Res 2011; 108: 871–883.

3. Medeiros-Domingo A, Bhuiyan ZA, Tester DJ, Hofman N, Bikker H, van Tintelen JP, et al. The RYR2-encoded ryanodine receptor/calcium release channel in patients diagnosed previously with either catecholaminergic polymorphic ventricular tachycardia or genotype negative, exercise-induced long QT syndrome: A comprehensive open reading frame mutational analysis. J Am Coll Cardiol 2009; 54: 2065–2074.

4. Hayashi M, Denjoy I, Extramiana F, Maltret A, Buisson NR, Lupoglazoff JM, et al. Incidence and risk factors of arrhythmic events in catecholaminergic polymorphic ventricular tachycardia. Circulation 2009; 119: 2426–2434.

5. Kawamura M, Ohno S, Naiki N, Nagaoka I, Dochi K, Wang Q, et al. Genetic background of catecholaminergic polymorphic ventricular tachycardia in Japan. Circ J 2013; 77: 1705–1713.

6. Marks AR. Calcium cycling proteins and heart failure: Mechanisms and therapeutics. J Clin Invest 2013; 123: 46–52.

7. Tung CC, Lobo PA, Kimlicka L, Van Petegem F. The aminoterminal disease hotspot of ryanodine receptors forms a cytoplasmic vestibule. Nature 2010; 468: 585–588.

8. Yuchi Z, Lau K, Petegem FV. Disease mutations in the ryanodine receptor central region: Crystal structures of a phosphorylation hot spot domain. Structure 2012; 20: 1–11.

9. Kobayashi S, Susa T, Tanaka T, Murakami W, Fukuoka S, Okada S, et al. Low-dose \( \beta \)-blocker in combination with milrinone safely improves cardiac function and eliminates pulse alternans in patients with acute decompensated heart failure. Circ J 2012; 76: 1646–1653.

10. Swan H, Laitinen P, Kontula K, Torvonen L. Calcium channel antagonism reduces exercise-induced ventricular arrhythmias in catecholaminergic polymorphic ventricular tachycardia patients with RyR2 mutations. J Cardiovasc Electrophysiol 2005; 16: 162–166.

11. Van der Werf C, Kannankeril PJ, Sacher F, Krahn AD, Viskin S, Leenhardt A, et al. Flecainide therapy reduces exercise-induced ventricular arrhythmias in patients with catecholaminergic polymorphic ventricular tachycardia. J Am Coll Cardiol 2011; 57: 2244–2254.

12. Watanabe H, Chopra N, Laver D, Hwang HS, Davies SS, Roach DE, et al. Flecainide prevents catecholaminergic polymorphic ventricular tachycardia in mice and humans. Nat Med 2009; 15: 380–383.

13. Liu N, Denegri M, Ruan Y, Avelino-Cruz JE, Perissi A, Negri S, et al. Flecainide exerts an antiarrhythmic effect in a mouse model of catecholaminergic polymorphic ventricular tachycardia by increasing the threshold for triggered activity [Short Communication]. Circ Res 2011; 109: 291–295.

14. Makanjee B, Gollob MH, Klein GI, Krahn AD. Ten-year follow-up of cardiac sympatheticectomy in a young woman with catecholaminergic polymorphic ventricular tachycardia and an implantable cardioverter defibrillator. J Cardiovasc Electrophysiol 2009; 20: 1167–1169.

15. Kobayashi S, Yano M, Uchinoumi H, Suotomi T, Susa T, Oto M, et al. Dantrolene, a therapeutic agent for malignant hyperthermia, inhibits catecholaminergic polymorphic ventricular tachycardia in a RyR2\textsuperscript{R2474S/+} knock-in mouse model. Circ J 2010; 74: 2579–2584.

16. Zhou Q, Xiao J, Jiang D, Wang R, Vembiayan K, Wang A, et al. Carvedilol and its new analogs suppress arrhythmogenic store overload-induced Ca\textsuperscript{2+} release. Nat Med 2011; 17: 1003–1009.

17. Hwang HS, Hasdemir C, Laver D, Mehra D, Turhan K, Faggioni M, et al. Antiarrhythmic drugs in catecholaminergic polymorphic ventricular tachycardia inhibition of cardiac Ca\textsuperscript{2+} release channels (RyR2) determines efficacy of Class I antiarrhythmic drugs in catecholaminergic polymorphic ventricular tachycardia. Circ Arrhythm Electrophysiol 2011; 4: 128–135.