Fever-triggered ventricular arrhythmias in Brugada syndrome and type 2 long-QT syndrome

A.S. Amin, C.A. Klemens, A.O. Verkerk, P.G. Meregalli, A. Asghari-Roodsari, J.M.T. de Bakker, C.T. January, A.A.M. Wilde, H.L. Tan

The risk for lethal ventricular arrhythmias is increased in individuals who carry mutations in genes that encode cardiac ion channels. Loss-of-function mutations in *SCN5A*, the gene encoding the cardiac sodium channel, are linked to Brugada syndrome (BrS). Arrhythmias in BrS are often preceded by coved-type ST-segment elevation in the right-precordial leads V1 and V2. Loss-of-function mutations in *KCNH2*, the gene encoding the cardiac ion channel that is respon-

sible for the rapidly activating delayed rectifying potassium current, are linked to long-QT syndrome type 2 (LQT-2). LQT-2 is characterised by delayed cardiac repolarisation and rate-corrected QT interval (QTc) prolongation. Here, we report that the risk for ventricular arrhythmias in BrS and LQT-2 is further increased during fever. Moreover, we demonstrate that fever may aggravate coved-type ST-segment elevation in BrS, and cause QTc lengthening in LQT-2. Finally, we describe molecular mechanisms that may underlie the proarrhythmic effects of fever in BrS and LQT-2. (Neth Heart J 2010;18:165-9.)

Keywords: Brugada Syndrome; Long QT Syndrome; Tachycardia, Ventricular; Fever

The risk for sudden death due to ventricular arrhythmias is increased in individuals who carry mutations in genes that encode cardiac ion channels and associated proteins. Inherited or *de novo* mutations in *SCN5A*, the gene that encodes the cardiac sodium channel, are linked to Brugada syndrome (BrS), a disease characterised by signs of conduction slowing on the ECG (prolonged PR and QRS intervals), and increased incidence of ventricular arrhythmias that are preceded by coved-type ST-segment elevation in the precordial leads V1 and V2.¹ BrS-linked *SCN5A* mutations exert their proarrhythmic effects by causing sodium channel loss-of-function (i.e., reducing cardiac sodium current [I_{Na}]) by disrupting intracellular trafficking of mutant channel proteins from the endoplasmic reticulum to the sarcolemma, forming channels that conduct no or very small I_{Na} , or altering the opening and closing (gating) properties of mutant channels in such a manner that the channels are less frequently open.^{2,3} Anecdotal reports have suggested that fever may aggravate

A.S. Amin

A.O. Verkerk

Heart Failure Research Centre, Department of Experimental Cardiology, Academic Medical Center, University of Amsterdam, Amsterdam, the Netherlands

C.A. Klemens

A. Asghari-Roodsari

J.M.T. de Bakker

Heart Failure Research Centre, Department of Experimental Cardiology, Academic Medical Center, University of Amsterdam, Amsterdam, and Interuniversity Cardiology Institute Netherlands

P.G. Meregalli

Department of Cardiology, Academic Medical Center, University of Amsterdam, Amsterdam, the Netherlands

C.T. January

Departments of Medicine and Physiology, Cellular and Molecular Arrhythmia Research Program, University of Wisconsin, Madison, Wisconsin, USA

A.A.M. Wilde

H.L. Tan

Heart Failure Research Centre, Department of Experimental Cardiology, and Department of Cardiology, Academic Medical Center, University of Amsterdam, Amsterdam, the Netherlands

Correspondence to: H.L. Tan

Department of Cardiology, Academic Medical Center, PO Box 22660, 1100 DD Amsterdam, the Netherlands
E-mail: h.l.tan@amc.nl

the typical ECG changes and increase the risk for sudden death in BrS.^{4,5} However, the association between fever and the risk for ventricular arrhythmias and preceding ECG changes has not been studied systematically. Recently, we first investigated this association in index patients who were diagnosed with BrS at our institution.⁶ In this retrospective study, we assessed the prevalence of fever-triggered ventricular tachycardia, ventricular fibrillation, or sudden death in 111 BrS patients, and compared these data with 41 control participants who were admitted after out-of-hospital cardiac arrest due to ventricular tachycardia or ventricular fibrillation. We found that, compared with control participants, BrS patients experienced cardiac arrest significantly more often during fever. Additionally, we analysed fever-induced ECG changes in 24 BrS patients for whom 12-lead ECGs during both fever and normothermia were available, and compared the data with ECG changes in ten control participants who were admitted for noncardiac reasons. We found that fever, irrespective of the cause, induced additional prolongation of PR and QRS intervals, and aggravated the coved-type ST-segment elevation in leads V1 and V2 in BrS patients. In contrast, the PR interval shortened, and the QRS interval and the ST-segment amplitude in leads V1 and V2 did not change during fever in control participants (figure 1A and B). We also investigated whether the use of antipyretic drugs during fever affected the prevalence of cardiac arrest in BrS patients. Indeed, a larger proportion of patients who had not used paracetamol (acetaminophen) experienced cardiac arrest during fever than those who had used acetaminophen. Thus, this systematic study indicated that fever increases the risk for ventricular arrhythmias and aggravates the typical ECG changes in BrS patients.

Despite the clinical evidence, the molecular mechanism underlying fever-triggered ventricular arrhythmias in BrS is not well understood. Cellular electrophysiological studies suggest that *SCN5A* mutations impair cardiac sodium channel gating more severely at higher temperatures, leading to further I_{Na} reduction during fever.⁷⁻⁹ Gating results from transitions of channel proteins between distinct conformational states. Upon membrane depolarisation, sodium channels change from their closed state to their open state (activation), thereby allowing influx of sodium ions into the cell. This causes more membrane depolarisation and triggers the cardiac action potential. Within milliseconds thereafter, sodium influx declines as sodium channels inactivate. Inactivation comprises multiple conformational states (fast, intermediate, and slow inactivation). To be able to open again for the next action potential, the channels must first return from the inactivated to the closed state (recovery from inactivation). This occurs during the diastolic interval.¹⁰ We and others have demonstrated that some *SCN5A* mutations cause sodium channels to enter their slow inactivation state more easily at higher temperatures (i.e., enhanced slow inactivation), thereby causing further I_{Na} reduction (figure 1C).^{7,8} Further I_{Na} reduction at higher temperatures is believed to cause additional conduction slowing, which corresponds

with the prolongation of PR and QRS intervals in BrS patients during fever.^{6,11,12} Interestingly, increased dysfunction of mutant sodium channels at higher temperatures is not specific for *SCN5A* mutations, since mutations in genes encoding other organ-specific sodium channels are also related to diseases with temperature-dependent symptoms. Mutations in *SCN1A*, the gene that encodes the brain sodium channel, is linked to generalised epilepsy with fever-triggered seizures.¹³ Mutations in *SCN9A*, the gene that encodes a sodium channel predominantly expressed in sensory and sympathetic neurones, is linked to primary erythralgia, a disease characterised by intermittent heat-triggered burning pain and skin redness in the extremities.¹⁴

Importantly, sinus tachycardia may also contribute to the development of arrhythmias during fever. During sinus tachycardia, the time between two consecutive depolarisations (i.e., heart beats) may become too short for mutant channels to full recover from inactivation. This may result in the accumulation of mutant channels in the slow inactivation state at fast heart rates, leading to further I_{Na} reduction. This hypothesis is supported by the aggravation of BrS-linked ECG changes in BrS patients during exercise-related tachycardia, and in right ventricular tissue preparations at higher stimulation frequencies.^{15,16}

Finally, fever-induced ventricular arrhythmias are not limited to BrS. Recently, we demonstrated that individuals with type 2 long-QT syndrome (LQT-2) may be at increased risk for arrhythmias during fever.¹⁷ LQT-2 is caused by loss-of-function mutations in *KCNH2* (formerly called *HERG*), the gene that encodes the channel carrying the rapidly activating delayed rectifying potassium current (I_{Kr}). I_{Kr} plays an important role in ventricular repolarisation, and I_{Kr} reduction delays ventricular repolarisation to cause action potential and QT-interval prolongation. We reported two related patients (father and son) with LQT-2 due to the heterozygous A558P *HERG* missense mutation who repeatedly experienced syncope and ventricular arrhythmias during fever. ECG analysis showed QTc lengthening in both LQT-2 patients. In contrast, QTc intervals in nine control participants shortened during febrile temperatures (figure 2A). When expressed in heterologous expression systems (HEK-293 cells), A558P mutant proteins displayed loss-of-function due to deficient intracellular trafficking (figure 2B). Because patients were heterozygous, carrying both normal and mutant allele, we co-expressed wild-type (WT) *HERG* proteins with A558P mutant proteins (WT+A558P), and found that A558P disrupts the intracellular trafficking of WT proteins (i.e., dominant-negative effect), causing 65% current reduction (figure 2B). Moreover, A558P reduced the normal increase of WT current at higher temperatures. In other words, the WT+A558P current failed to increase to the same extent as the WT current and caused larger reduction in current density at higher temperatures. A similar temperature-dependent phenotype was seen for co-expression of WT proteins with the F640V *HERG* mutation (figure 2C and D). Similar to A558P, F640V is linked

Figure 1. A) Fever-induced ECG changes in leads V1 and V2 of a Brugada syndrome patient. B) ECG variables during normothermia and fever for Brugada syndrome patients and control participants. * $p < 0.05$ versus baseline. C) Slow inactivation in wild-type (WT) and mutant (D1714G) cardiac sodium channels at 21°C and 36°C. Using the patch-clamp technique, HEK-293 cells expressing WT or mutant SCN5A were stimulated with an initial conditioning pre-pulse to -20 mV (P1) of variable duration ranging between 1 and 1000 ms, followed by a 20 ms (21°C) or 2 ms (36°C) step to -120 mV to allow channels to recover from fast inactivation, and a final test pulse to -20 mV (P2) to assess the fraction of channels that had exited slow inactivation and were available for reactivation (inset). This fraction was obtained by dividing the current during P2 by the current during P1, and plotted versus the duration of P1. * $p < 0.05$ compared with 21°C. Slow inactivation is enhanced in D1714G channels at 36°C, but not at 21°C (adapted from Amin et al.).⁸

to LQT-2, displays a trafficking-deficient phenotype, and exerts a dominant-negative effect.¹⁸ We postulated that the reduced increase in the current density in WT-mutant

co-assembled channels at higher temperatures may underlie QTc lengthening and the development of ventricular arrhythmias during fever. Indeed, I_{Kr} reduction at febrile

temperatures has been shown to prolong action potential duration in Purkinje fibre preparations, thereby increasing transmural differences in action potential duration, and facilitating the occurrence of early afterdepolarisations (which are believed to result from reactivation of L-type Ca^{2+} channels during phase 3 of the cardiac action potential).^{19,20} Early afterdepolarisations are believed to initiate ventricular arrhythmias in LQT-2.²¹

In summary, we have shown that individuals with BrS or LQT-2 may be at increased risk for ventricular arrhythmias during fever. Accordingly, we demonstrated that fever may aggravate the coved-type ST-segment elevation in leads V1 and V2 that often precedes arrhythmias in BrS, and may cause QTc lengthening in LQT-2. The possible underlying mechanisms involve the effects of fever on the function of mutant ion channels. Nevertheless, fever-triggered arrhythmias may not only be attributed to further dysfunction of mutant ion channels, since they may also occur in individuals without mutations in ion channel encoding genes.²²⁻²⁴ Importantly, early treatment of fever with antipyretic drugs may lower the risk for ventricular arrhythmias during fever in BrS, and perhaps also in LQT-2. Moreover, although evidence is lacking, vaccination against seasonal flu viruses, and in particular the 2009 H1N1 virus, may lower arrhythmia risk by preventing severe febrile episodes due to viral infections. Future research will focus on the *in vivo* effects of fever and its various aspects (e.g., higher body temperature, sinus tachycardia, and inflammatory cells and cytokines) on cardiac electrophysiology in transgenic mouse models carrying mutations in a specific gene of interest, and in individuals without mutations in ion channel encoding genes but with common acquired diseases (e.g., myocardial infarction, and congestive heart failure).

Acknowledgments

This study was supported by the Netherlands Organisation for Scientific Research grants NWO ZonMW-VICI 918-86-616 (Hanno L. Tan), and NWO 902-16-193 (Arthur A.M. Wilde), the Netherlands Heart Foundation grant NHS 2003T302 (Arthur A.M. Wilde), and the National Heart, Lung, and Blood Institute grant R01 HL60723 (Craig T. January).

References

- 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.
- Amin AS, Asghari-Roodsari A, Tan HL. Cardiac sodium channelopathies. *Pflügers Arch-Eur J Physiol*. 2009. DOI 10.1007/s00424-009-0761-0.
- January CT, Riddle JM. Early afterdepolarizations: Mechanism of induction and block. A role for L-type Ca^{++} current. *Circ Res*. 1989;64:977-90.
- Tan HL, Meregalli PG. Lethal ECG changes hidden by therapeutic hypothermia. *Lancet*. 2007;369:78.
- Patruno N, Pontillo D, Achilli A, Ruggeri G, Critelli G. Electrocardiographic pattern of Brugada syndrome disclosed by a febrile illness: clinical and therapeutic implications. *Europace*. 2003;5:251-5.
- Amin AS, Meregalli PG, Bardai A, Wilde AA, Tan HL. Fever increases the risk for cardiac arrest in the Brugada syndrome. *Ann Intern Med*. 2008;149:216-8.
- Dumaine R, Towbin JA, Brugada P, Vatta M, Nesterenko DV, Nesterenko VV, et al. Ionic mechanisms responsible for the electrocardiographic phenotype of the Brugada syndrome are temperature dependent. *Circ Res*. 1999;85:803-9.
- Amin AS, Verkerk AO, Bhuiyan ZA, Wilde AAM, Tan HL. Novel Brugada syndrome-causing mutation in ion-conducting pore of cardiac Na^+ channel does not affect ion selectivity properties. *Acta Physiol Scand*. 2005;185:291-301.
- Wang DW, Makita N, Kitabatake A, Balser JR, George AL Jr. Enhanced Na^+ channel intermediate inactivation in Brugada syndrome. *Circ Res*. 2000;87:E37-43.
- Balser JR. The cardiac sodium channel: gating function and molecular pharmacology. *J Mol Cell Cardiol*. 2001;33:599-613.
- Tukkie R, Sogaard P, Vleugels J, de Groot IK, Wilde AA, Tan HL. Delay in right ventricular activation contributes to Brugada syndrome. *Circulation*. 2004;109:1272-7.
- Meregalli PG, Wilde AA, Tan HL. Pathophysiological mechanisms of Brugada syndrome: depolarization disorder, repolarization disorder, or more? *Cardiovasc Res*. 2005;67:367-78.
- Claes L, Del-Favero J, Ceulemans B, Lagae L, Van Broeckhoven C, De Jonghe P. De novo mutations in the sodium-channel gene *SCN1A* cause severe myoclonic epilepsy of infancy. *Am J Hum Genet*. 2001;68:1327-32.
- Yang Y, Wang Y, Li S, Xu Z, Li H, Ma L, et al. Mutations in *SCN9A*, encoding a sodium channel alpha subunit, in patients with primary erythromalgia. *J Med Genet*. 2004;41:171-4.
- Amin AS, de Groot EA, Ruijter JM, Wilde AA, Tan HL. Exercise-induced ECG changes in Brugada syndrome. *Circ Arrhythm Electrophysiol*. 2009;2:531-9.
- Coronel R, Casini S, Koopmann TT, Wilms-Schopman FJ, Verkerk AO, de Groot JR, et al. Right ventricular fibrosis and conduction delay in a patient with clinical signs of Brugada syndrome: a combined electrophysiological, genetic, histopathologic, and computational study. *Circulation*. 2005;112:2769-77.
- Amin AS, Herfst LJ, Delisle BP, Klemens CA, Rook MB, Bezzina CR, et al. Fever-induced QTc prolongation and ventricular arrhythmias in individuals with type 2 congenital long QT syndrome. *J Clin Invest*. 2008;118:2552-61.
- Anderson CL, Delisle BP, Anson BD, Kilby JA, Will ML, Tester DJ, et al. Most LQT2 mutations reduce Kv11.1 (hERG) current by a class 2 (trafficking-deficient) mechanism. *Circulation*. 2006;113:365-73.
- Burashnikov A, Shimizu W, Antzelevitch C. Fever accentuates transmural dispersion of repolarization and facilitates development of early afterdepolarizations and torsade de pointes under long-QT Conditions. *Circ Arrhythm Electrophysiol*. 2008;1:202-8.
- Delisle BP, Anson B, Rajamani S, January CT. Biology of cardiac arrhythmias: Ion channel protein trafficking. *Circ Res*. 2004;94:1418-28.
- Yan GX, Wu Y, Liu T, Wang J, Marinichak RA, Kowey PR. Phase 2 early afterdepolarization as a trigger of polymorphic ventricular tachycardia in acquired long-QT syndrome: direct evidence from intracellular recordings in the intact left ventricular wall. *Circulation*. 2001;103:2851-6.
- Pasquie JL, Sanders P, Hocini M, Hsu LF, Scavee C, Jais P, et al. Fever as a precipitant of idiopathic ventricular fibrillation in patients with normal hearts. *J Cardiovasc Electrophysiol*. 2004;15:1271-6.
- D'Aloia A, Faggiano P, Brentana L, Boldini A, Curnis A, Bontempi L, et al. Recurrent ventricular fibrillation during a febrile illness and hyperthermia in a patient with dilated cardiomyopathy and automatic implantable cardioverter defibrillator. An example of reversible electrical storm. *Int J Cardiol*. 2005;103:207-8.
- Burrell C, Reddy S, Haywood G, Cunningham R. Cardiac arrest associated with febrile illness due to U.K. acquired *Cyclospora cayentanensis*. *J Infect*. 2007;54:e13-5.

Figure 2. A) QTc durations in LQT-2 patients and control participants in relation to body temperature. Solid lines represent regression lines based on the common slopes. The common slope of the fit through LQT-2 patients differs significantly from that of control participants. B) Average current densities measured at 23°C from HEK-293 cells expressing wild-type (WT), A558P, or WT+A558P HERG. * $p < 0.05$ compared with WT. C) Average current densities measured at 23°C, 35°C, and 40°C from HEK-293 cells expressing WT, WT+A558P, or WT+F640V HERG. The temperature-dependent increase in current density is smaller for WT+A558P and WT+F640V channels compared with WT channels. D) Average differences in current densities between WT and WT+A558P or WT+F640V at 23°C, 35°C, and 40°C. Differences in current densities increase with rising temperature. * $p < 0.05$ compared with 23°C; # $p < 0.05$ compared with 35°C (adapted from Amin et al.).¹⁷