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REVIEW

Genetic and Clinical Advances in Congenital Long QT Syndrome

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Congenital long QT syndrome (LQTS) is an inherited arrhythmia syndrome characterized by a prolonged QT interval on the 12-lead ECG, torsades de pointes and a higher chance of sudden cardiac death. LQTS segregates in a Mendelian fashion, which includes Romano-Ward syndrome with an autosomal dominant pattern as well as a rare autosomal recessive pattern (Jervell and Lange-Nielsen syndrome). Since 1957 when Jervell and Lange-Nielsen reported the first familial LQTS with congenital deafness, progress in understanding the genetic and electrophysiological mechanisms of LQTS has tremendously improved diagnostic methods and treatments. In the meantime, it has become evident that LQTS may not always be explained by a single gene mutation, but seems to follow a more complex genetic model intertwined with genetic common polymorphisms that have a mild to moderate effect on disease expression. In this review, we summarize the characteristics of LQTS (mainly LQT1–3) and briefly describe the most recent advances in LQTS clinical diagnostics as well as genetics. (*Circ J* 2014; **78:** 2827–2833)

Key Words: Beta-blockers; Diagnosis; Ion channels; Long QT syndrome; Torsade de pointes

Since the first description of a LQTS family in 1957,[1] tremendous progress in understanding its pathogenesis, diagnosis and treatments has been achieved. This review will describe the recent update of the diagnostic scoring system in 2011, the expert consensus statement published in 2013 and new challenges in discovering genotype-phenotype relationship in LQTS. Because of limited space, our focus is mainly on the general concept of the most frequently encountered Romano-Ward syndrome (RWS; LQTS types 1–3). Details of minor LQTS subtypes are to be found elsewhere.[2–4]

Clinical Characteristics

The prevalence of congenital LQTS is reported to be approximately 1/2,000.[5] Syncope is generally the most commonly encountered first episode in LQTS patients, and aborted cardiac arrest/sudden cardiac death (ACA/SCD) is rare (1–3%).[6] Of the patients who eventually become symptomatic, 50% experience their first cardiac event by the age of 12, and 90% by the age of 40.[7] LQTS is also known as an etiology of sudden infant death syndrome (SIDS)[8] and in approximately 10% of SIDS cases the infant carried a mutation in a LQTS-causing gene.[9]

The most frequent LQTS subtypes are type 1 (LQT1), type 2 (LQT2) and type 3 (LQT3).[10] The subdivision is based on the underlying genetic substrate, with the potassium channel genes, *KCNQ1*, *KCNH2*, and the sodium channel gene, *SCN5A*, as the involved genes. Specific triggers of symptoms are known in these major LQTS subtypes. For example in LQT1, cardiac events occur during physical exertion or emotional stress, typically during swimming,[11] whereas auditory stimulation such as an alarm clock or a telephone bell is a typical trigger for arrhythmic events in LQT2.[12] Emotional stress, as well as the postpartum period, are also frequently observed triggers of arrhythmia in LQT2.[11,13] More recently, a history of epilepsy has been reported to be more common with LQT2 (39%) than with other subtypes of LQTS (10%, P<0.001),[14] possibly because *KCNH2* is expressed in the brain as well.[14] Obviously in such cases, the differential diagnosis of LQTS and epilepsy is important for treatment, because LQTS is often mistreated as epilepsy because of generalized seizures secondary to torsades de pointes. In LQT3, symptoms are most frequently observed during rest or at night.[11,15]

Diagnosis

Table 1 shows the LQTS diagnostic scoring system (updated in 2011), which includes symptoms, family history and ECG findings.[16] Patients with a Schwartz score ≥3.5 points in the absence of a secondary cause for QT prolongation are diagnosed as LQTS.[17] Typical LQTS cases can be readily diagnosed with this scoring system, whereas latent LQTS patients with normal QTc at rest, found in 36% of LQT1, 19% of LQT2 and 10% of LQT3, respectively,[18] may show a non-diagnostic

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Table 1. Schwartz Score (Updated in 2011)[16]

	Points
ECG findings	
QTc	
>480 ms	3
460–470 ms	2
450 (male) ms	1
4-min recovery QTc after exercise test ≥480 ms	1
Torsades de pointes	2
T-wave alternans	1
Notched T wave in 3 leads	1
Low heart rate for age	0.5
Clinical history	
Syncope	
With stress	2
Without stress	1
Congenital deafness	0.5
Family history	
A. Family members with definite LQTS	1
B. Unexplained sudden cardiac death <age 30 among immediate family members	0.5

LQTS diagnostic criteria. QTc is calculated by Bazett's formula where $QTc = QT/\sqrt{RR}$. Low heart rate for age means resting heart rate below the 2nd percentile for age.

score of 1–3 points. In such cases, serial ECGs, 24-h Holter recordings, and an exercise or epinephrine test are recommended to reveal subclinical QT prolongation.[19–21]

With regard to the exercise test, the QT interval during the recovery phase is valuable in LQTS diagnostics.[22,23] In 69 relatives of LQT1 and LQT2 probands who showed borderline to normal QTc at rest, Sy et al reported that a 4-min recovery QTc ≥445 ms discriminated LQTS gene carriers from non-carriers, and a 4-min recovery ≥480 ms showed 100% specificity.[22] Horner et al reported from exercise testing of 243 patients that paradoxical QTc prolongation during the recovery phase (QTc ≥460 ms) distinguished LQTS, particularly LQT1, and ΔQTc ≥30 ms (QTc 3-min recovery minus the baseline supine QTc) showed a high sensitivity and specificity (83% and 93%, respectively).[23] Importantly, both studies reported that exercise testing showed satisfactory results in distinguishing LQTS cases from innocent ones while on β-blockers, suggesting no need for a β-blocker washout.[22,23] As another diagnostic method in LQTS, brisk movement from supine to upright has been reported.[24] In that study, QTc was significantly increased by 89±47 ms in the LQTS group compared with 50±30 ms in the control group during maximal sinus tachycardia induced by standing up.[24]

To note, the QT interval measurement can vary according to physician and LQTS expertise.[25] In addition, accurate measurement of the QTc interval at high heart rate (HR) is particularly challenging. In the case of children, by analyzing ECGs from a school-based screening program, Yoshinaga et al managed to appropriately diagnose genotype-positive LQTS cases with a QTc cut-off value (Bazett's formula) ≥450 ms for HR <75 beats/min and ≥500 ms for HR ≥75 beats/min (94% in screened children), although it was not shown how many genotype-positive patients were missed by the use of these cut-off values.[26] In any case, an expert eye on the QT interval (and ST-T morphology) remains a cornerstone of LQTS diagnostics.

In addition to the diagnostic criteria just mentioned, T wave morphology may help differentiate LQT1–3.[27] Typically, a broad T wave is observed in LQT1 (Figure A), a biphasic T wave in LQT2 (Figure B) and a late-appearing T wave in LQT3, which has a narrow and tall shape, and appears at the very end of the QT interval (Figure C).[27]

Furthermore, genotyping is certainly important in the diagnosis and treatment of LQTS.[17] Currently in Asia, LQTS genetic testing (candidate gene approach) is reimbursed in Korea (by Korean NIH for 13 LQTS genes) and in Japan (70% covered by the government public health insurance), but it remains at research level at least in Bangladesh, China, India, Taiwan and Thailand because of either high costs or lack of facilities for broad use (personal communication). Currently, in the era of next-generation sequencing, technological advances enable us to screen many more genes through disease-associated gene panels or even whole exome sequencing. However, with these methods, there are numerous rare "variants of unknown significance" reported with unknown risk associated with disease. Such variants remain elusive and await further research to establish genotype-phenotype relationships before they can used in the clinical setting.

Electrophysiological and Genetic Abnormalities in LQTS

The QT interval reflects repolarization of the ventricular action potentials orchestrated by various cardiac ion currents, including sodium, calcium and potassium currents.[28] When a decreased potassium current (loss-of-function) or increased sodium or calcium current (gain-of-function) is caused by a mutation in an ion channel or axillary protein, a prolongation of the action potential duration occurs, which manifests on the 12-lead ECG as QT prolongation.

In 1995 and 1996, causal gene mutations of familial LQTS in 3 genes (*KCNQ1*, *KCNH2* and *SCN5A*) were discovered.[28–30] To date, mutations in 15 different genes have been reported in LQTS (Table 2),[31–41] but *KCNQ1*, *KCNH2* and *SCN5A* remain numerically the major genes in LQTS and comprise more than 90% of genotype-positive cases of LQTS.[10] A causal mutation is found in approximately 75% of LQTS patients with a Schwartz score ≥4,[42] but the genetic background of the other 25% of patients remain elusive.

In approximately 85% of genotype-positive cases of LQTS, the patient carries a mutation inherited from one of the parents and in the remaining 15% a de novo mutation is pertinent.[43] In genotyped LQTS patients, approximately 50% have no lifetime symptoms, and 10–50% of such patients show no apparent QT prolongation.[44–46] Compound mutations (ie, ≥2 mutations) are found in 10% of genotype-positive patients[47] and the clinical manifestation of disease in such patients is often more severe.[48,49]

As for minor LQTS subtypes with severe clinical phenotypes, mutations in *KCNQ1* or *KCNE1* present as homozygous or compound heterozygous mutations in Jervell and Lange-Nielsen syndrome (J-LNS; Table 2).[50,51] *KCNQ1* mutations are much more prevalent (90%) than *KCNE1* mutations (10%).[52] Calmodulin de novo mutations were reported in infant cases of recurrent cardiac arrest by means of exome sequencing in the parents-child trio.[41]

Genetic Modifiers of LQTS

Since the beginning of research into the genetic background in LQTS, linkage analysis and the candidate gene approach

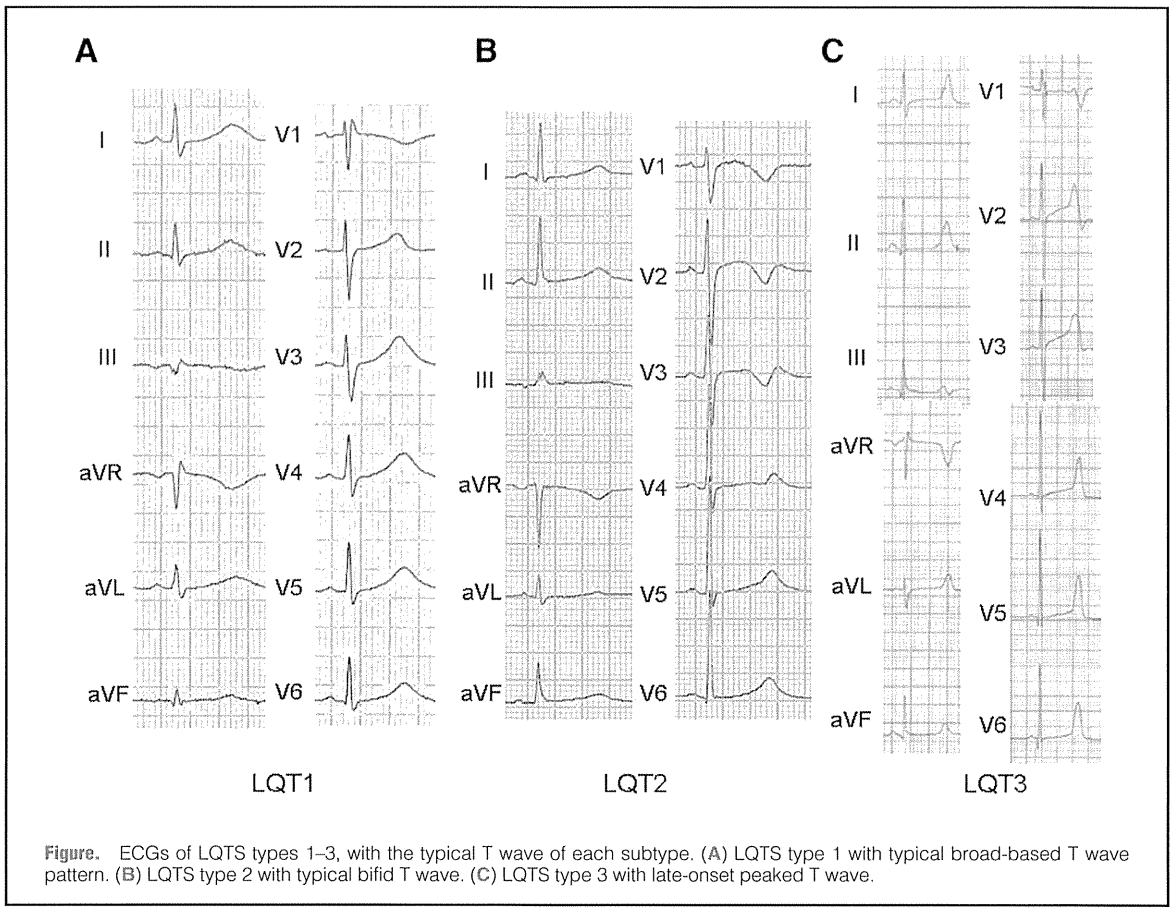

Figure. ECGs of LQTS types 1–3, with the typical T wave of each subtype. (**A**) LQTS type 1 with typical broad-based T wave pattern. (**B**) LQTS type 2 with typical bifid T wave. (**C**) LQTS type 3 with late-onset peaked T wave.

Table 2. Congenital LQTS Genes and Affected Ion Currents

LQTS type	Gene	Protein	Current	Frequency (%)
Romano-Ward				
LQT1	*KCNQ1*	Kv7.1	↓ I_{Ks}	40–55
LQT2	*KCNH2*	Kv11.1	↓ I_{Kr}	30–45
LQT3	*SCN5A*	Nav1.5	↑ I_{Na}	5–10
LQT4	*ANKB*	Ankyrin	↓ Coordination of Ncx, Na/K ATPase	Rare
LQT5	*KCNE1*	MinK	↓ I_{Ks}	Rare
LQT6	*KCNE2*	MiRP1	↓ I_{Kr}	Rare
LQT7	*KCNJ2*	Kir2.1	↓ I_{K1}	Rare
LQT8	*CACNA1C*	Cav1.2	↑ I_{Ca}	Rare
LQT9	*CAV3*	Caveolin 3	↑ I_{Na}	Rare
LQT10	*SCN4B*	Sodium channel β4-subunit	↑ I_{Na}	Very rare
LQT11	*AKAP9*	Yotiao	↓ I_{Ks}	Very rare
LQT12	*SNTA1*	Syntrophin-α1	↑ I_{Na}	Very rare
LQT13	*KCNJ5*	Kir3.4	↓ $I_{K\text{-}Ach}$	Very rare
LQT14	*CALM1*	Calmodulin 1	Dysfunctional Ca^{2+} signaling	Rare
LQT15	*CALM2*	Calmodulin 2		Rare
Jervell and Lange-Nielsen				
JLN1	*KCNQ1*	Kv7.1	↓ I_{Ks}	Rare
JLN2	*KCNE1*	MinK	↓ I_{Ks}	Rare

have been used, assuming LQTS to be a monogenic model. Later, it became evident from research into large LQTS families that LQTS actually shows incomplete penetrance with variable expressivity. In other words, different clinical phenotypes (sudden death, syncope, asymptomatic) are observed in family members carrying the same familial mutation, which may be caused by a more complex genetic model involving multiple genetic and environmental factors affecting disease development.

Modifier genes are common genetic variants found in more than 1% of affected individuals, which have an influence on susceptibility to disease. They have less effect on disease compared with a causal gene mutation, but by summing up the effects of such modifier genes, the disease severity varies. In LQTS, variants in the coding regions of LQTS-related genes such as *KCNE1* D85N,[53] *KCNH2* K897T,[54] and *SCN5A* H558R[55] are known to influence the QT interval. Single nucleotide polymorphisms (SNPs) in non-coding regions (intron, 3'UTR) of *KCNQ1* are also reported as an important QT-interval modifier.[56,57] Furthermore, genome-wide association studies have revealed SNPs in *NOS1AP* as a modulator of QT interval in LQTS patients,[58,59] as well as in the general population.[60] An effort to find any SNPs affecting QT interval of the Caucasian general population continues by larger international consortia.[61] This field is expected to expand to further understand the complexity of genotype-phenotype relationship in LQTS. However, it remains currently at the research level, which requires careful interpretation by experts.[62]

Risk Assessment of Cardiac Events

The well-established parameters of risk stratification in LQTS patients are prolonged QTc interval ≥500 ms and a history of syncope.[18] More detailed studies have revealed that the risk of a first cardiac event in males is higher in childhood before puberty, whereas in females, cardiac events occur during adolescence and the postpartum period.[63,64] In adults between 18 and 40 years of age, the risks of experiencing any cardiac event are a history of ≥1 cardiac events before age 18 years, female sex, longer QTc interval (≥440 ms) and LQT2.[65] For patients older than 40 years, LQT3 patients have significantly more cumulative lethal arrhythmic events (35%) than LQT1 (14%), LQT2 (24%) and genotype-negative patients (10%).[66,67] Importantly, a family history of SCD in a first-degree relative is not a significant risk for ACA/SCD.[68]

LQTS patients with a normal QTc interval (≤440 ms) carry a lower risk of ACA/SCD compared with LQTS patients with QT prolongation (QTc >440 ms), but still show >10-fold risk of fatal arrhythmia compared with genotype- and phenotype-negative family members.[69] Such "latent" cases of LQTS can be recapitulated by cellular electrophysiological studies and can give further insights to the mechanisms. For example, Wu et al used a heterozygous construct of *KCNQ1*-G269S in CHO and HEK cells and showed that this mutation located in segment 5 of I_{Ks} channel significantly disrupted upregulation of I_{Ks} currents in response to protein kinase A (PKA) stimulation, which fits with the clinical characteristics of patients carrying *KCNQ1*-G269S (borderline QT interval at rest and QT prolongation and syncope after exercise).[70]

More recent studies have shown that mutation location and type influence the variability of symptoms. In patients with LQT1, missense cytoplasmic-loop mutations have been reported to cause a longer QT interval at enrolment and an increased risk of ACA/SCD.[71] As a specific mutation in *KCNQ1*, A341V, more than other *KCNQ1* mutations (transmembrane or dominant-negative mutations), harbors the most severe phenotype.[72–74] In LQT2 patients, the risk of ACA/SCD is higher in females regardless of mutation site compared with males.[75] Among male subjects, patients with pore-loop mutations,[75] especially missense mutations,[76] show a higher incidence of ACA/SCD compared with non-pore-loop mutations.

Treatment

Drug Therapy

Beta-blocker therapy is the first choice for LQTS.[17] It dramatically decreases event rates, from 0.97 to 0.31 events per patient per year.[77] Recurrent events were often seen in patients with a history of ACA or in patients who are non-compliant with β-blocker therapy.[77] In LQT1 and LQT2, propranolol ($2–4\,mg \cdot kg^{-1} \cdot day^{-1}$) and nadolol ($1–1.5\,mg \cdot kg^{-1} \cdot day^{-1}$) have been shown to be much more effective than metoprolol in suppressing recurrent cardiac events.[78] Therefore, metoprolol should probably not be prescribed in symptomatic LQTS patients.[78,79] Atenolol, not included in the aforementioned study, appears to be less effective, according to a study performed in (only) 28 genotyped patients with a median follow-up of 46 months.[80]

Of the 3 major genotypes of LQTS, β-blocker therapy is extremely effective in LQT1 because of the prominent involvement of adrenergic stimulation in its pathogenesis.[11,70] Pure β-blockers are less effective in LQT2 than LQT1, possibly because of α_{1A} adrenoreceptor-mediated I_{Kr} reduction.[81] Preliminary data recently extracted from >400 LQT3 patients showed that β-blocker therapy is also protective in LQT3.[82]

As adjunctive therapy, some drugs are used in a genotype-specific manner. For example, oral K^+ supplements are considered especially in LQT2 patients because the underlying genetic defect is very sensitive to the serum potassium level.[83–85] This holds true particularly when the serum potassium level is low (eg, with diarrhea). In LQT3, mexiletine in addition to β-blocker therapy may be considered in patients with a specific mutation.[86] Of note, worsening of QT prolongation by mexiletine was reported in 1 individual with LQT3.[87] Therefore, it is important in LQT3 patients to re-check the QT interval after mexiletine administration when the serum mexiletine level reaches the therapeutic level.[79] Another drug, ranolazine, a FDA-approved anti-anginal agent, has recently drawn attention as an alternative choice to treat patients with LQTS because it blocks the late sodium current, $I_{Na\ late}$, (and to a minor extent the I_{Kr} and $I_{Ca,L}$ without proarrhythmic effects).[88] There is so far only 1 clinical report, which includes a small number of patients, on the short-term effectiveness of ranolazine in LQT3.[89]

Cardiac events in LQTS patients are generally well controlled with β-blocker therapy. However, patients with a history of ACA, symptomatic patients in the first year of life, and patients with J-LNS carrying *KCNQ1* mutations are at particularly high risk and therefore, special aids, including implantable cardioverter defibrillator (ICD) and left cardiac sympathetic denervation (LCSD) on top of β-blocker therapy may be deemed necessary because of the high recurrence rate of fatal arrhythmia.[77,90] Besides medical treatment, life-style advice for patients is important.[17]

LCSD

LCSD is a surgical procedure to ablate the lower two-thirds of the left stellate ganglion together with the thoracic ganglia T2–T4 to denervate cardiac sympathetic innervation to the heart.[91] It is currently used as an adjunctive therapy in symp-

tomatic patients who are refractory to β-blocker therapy. LCSD has been shown to reduce cardiac events significantly in LQTS with a rather high long-term cardiac-event-free survival (46%/5 years,[91] 59%/2 years[92]). In patients with a history of syncope, post-LCSD QTc <500 ms predicts efficacy of LCSD.[91] However, those with persistent QTc prolongation (≥500 ms) have a high chance of SCD and need to be protected with an ICD. A known complication of this procedure is Horner's syndrome, but in most cases, it is only transiently observed after the surgery and the patient recovers afterwards.[91]

ICD

Use of an ICD should be regarded as adjunctive therapy in LQTS. An ICD is recommended only for patients who have frequent syncopal episodes despite being on maximal doses of β-blocker (and eventually other additional pharmacological therapies) or at high risk of recurrent ACA/SCD,[17] such as patients who have a history of ACA, symptomatic infant cases (<1 year of age) or those with J-LNS.[52]

Conclusions

Research on LQTS in the past 2 decades has broadened our knowledge of the mechanism as well as genotype-specific therapeutic options. An ongoing challenge is LQTS with normal to borderline QTc at rest. In such cases, diagnostic tests to detect maladaptation of the QT interval to HR are clinically important. Indeed, the updated scoring system in 2011 now includes QT interval of a recovery phase after exercise. In LQTS genetics, next-generation sequencing enables us to detect SNPs in coding/non-coding regions of (LQTS-related) genes that modify the QT interval. Currently, analysis and interpretation of results by next-generation sequencing remains at the research level while advances in the understanding of genotype-phenotype relationships, including LQTS causal genes as well as SNPs, are expected to guide us further to genetically-guided personalized treatment in the future.

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Original Article

Genetic Characteristics of Children and Adolescents With Long-QT Syndrome Diagnosed by School-Based Electrocardiographic Screening Programs

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Background—A school-based electrocardiographic screening program has been developed in Japan. However, few data are available on the genetic characteristics of pediatric patients with long-QT syndrome who were diagnosed by this program.

Methods and Results—A total of 117 unrelated probands aged ≤18 years were the subjects who were referred to our centers for genetic testing. Of these, 69 subjects diagnosed by the program formed the screened group. A total of 48 subjects were included in the clinical group and were diagnosed with long-QT syndrome–related symptoms, familial study, or by chance. Mutations were classified as radical, of high probability of pathogenicity, or of uncertain significance. Two subjects in the clinical group died. Genotypes were identified in 50 (72%) and 23 (48%) of subjects in the screened and clinical groups, respectively. Of the *KCNQ1* or *KCNH2* mutations, 31 of 33 (94%) in the screened group and 14 of 15 (93%) in the clinical group were radical and of high probability of pathogenicity. Prevalence of symptoms before (9/69 versus 31/48; $P<0.0001$) and after (12/69 versus 17/48; $P=0.03$) diagnosis was significantly lower in the screened group when compared with that in the clinical group although the QTc values, family history of long-QT syndrome, sudden death, and follow-up periods were not different between the groups.

Conclusions—These data suggest that the screening program may be effective for early diagnosis of long-QT syndrome that may allow intervention before symptoms. In addition, screened patients should have follow-up equivalent to clinically identified patients. ***(Circ Arrhythm Electrophysiol.* 2014;7:107-112.)**

Key Words: diagnosis ■ genetic testing ■ QT interval electrocardiography ■ screening

Congenital long-QT syndrome (LQTS) is a genetic disorder characterized by delayed repolarization and by a long-QT interval on 12-lead ECGs. Although many patients do not have symptoms, the hallmark of the condition is syncope or sudden death because of torsade de pointes.[1,2] To date, 13 genes have been identified.[3,4] There have been many reports on the clinical and genetic backgrounds of patients with LQTS. However, these were mainly based on data collected from patients who had LQTS-related symptoms or familial studies and from combined adult and pediatric populations.[2,5–9]

Clinical Perspective on p 112

A nationwide school-based ECG screening program for heart diseases in first, seventh, and 10th graders in Japan has revealed children and adolescents with prolonged QT intervals. The prevalence of subjects with prolonged QT intervals was ≈1:1200 in the seventh grade.[10] Differences in clinical characteristics between patients who were screened by the program and those who visited hospitals with symptoms have been previously reported.[11] However, before this study, few data have been reported about the genetic characteristics of pediatric patients who were diagnosed by ECG screening programs and whose genetic testing was performed.[12–14] In addition, because of a lack of reports containing large numbers of patients who were screened alongside genetic testing, it is unclear whether screened subjects have similar mutations of a high possibility of pathogenesis to those who have LQTS-related symptoms.

From the genetic testing viewpoint, the few percentage background rate of the rare *KCNQ1* and *KCNH2* nonsynonymous single nucleotide variants among healthy individuals has lessened the ability to distinguish rare pathogenic mutations from similarly rare, yet presumably innocuous, variants.[15,16] Novel mutations have been found in every study,[17,18] but it is difficult to perform electrophysiological studies for

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each novel mutation except in large laboratories. Recently, an algorithm designed to guide the interpretation of genetic testing results for *KCNQ1* and *KCNH2* has been developed.[16]

Thus, the aim of the present study was to determine the genetic characteristics of pediatric patients with LQTS who were diagnosed by a school-based screening program and whose genetic testing was performed and to compare results with subjects who visited hospitals because of the presence of LQTS-related symptoms, familial history, or who were diagnosed by chance.

Methods

Study Population

The study population included 117 unrelated probands ≤18 years of age who were referred to the Department of Pediatrics, Kagoshima University Hospital, Japan, between November 1993 and March 2005 or to the National Hospital Organization Kagoshima Medical Center from April 2005 to December 2012 for genetic evaluation. The population included 69 subjects who were screened by a school-based ECG screening program (Table 1). In the present study, LQTS-related symptoms were defined as syncope, aborted cardiac arrest, or sudden cardiac death at <30 years old. Subjects were divided into 2 groups on the basis of index events: subjects who were diagnosed by the school-based ECG screening program (screened group) and those who visited hospitals because of the presence of symptoms and family history or who were diagnosed by chance (clinical group; Table 1).

Diagnosis of LQTS and Screening of QT Intervals in the School-based ECG Screening Program

The present study was a retrospective study, and diagnosis of LQTS and screening for prolonged QT intervals was based on the judgment of the chief medical doctors in each hospital or doctors who participated in the program in each area. Many Japanese cardiologists use a scoring system published in 1993[19] and recently[1] for the final diagnosis of LQTS. To screen subjects with prolonged QT intervals in the program, the Japanese Society of Pediatric Cardiology and Cardiac Surgery recommended that children and adolescents be screened when they have a QTc value, using Bazett formula, of ≥450 ms at a heart rate of <75 beats per minute or a QTc≥500 ms at a hours of ≥75 beats per minute.[20] Bazett formula overcorrects the QT interval at high heart rates. Pediatric cardiologists who participated in the program used age- and sex-specific criteria using an exponential formula ($QT/RR^{0.31}$)[21] or Fridericia formula.[22] In the screening program, cardiologists use computer-based QTc values as a reference because all ECG machines used in Japan are generally equipped with a function for automated measurement of QT intervals. However, manual measurement using the tangent method is usually applied to obtain QT intervals in Japan.[22,23]

Table 1. Characteristics of Probands

Subjects	Screened Group	Clinical Group	*P* Value
No. of subjects	69	48	...
Age at diagnosis*	10.4±3.4	7.4±6.0	0.04
Age at diagnosis (median and range)	12.2 (6.2–18.8)	8.9 (0–17.2)	...
Sex (men/women)	36/33	27/21	0.66
Mean QT interval, ms*	466±51	442±83	0.09
Mean RR interval, ms*	887±170	802±261	0.09
QTc (Bazett), $ms^{1/2}$*	496±40	502±52	0.84
History of symptoms†	9 (13%)	31 (65%)	<0.0001
Syncope	9	28	...
Aborted cardiac arrest	0	7‡	...
Family history of long-QT syndrome†	27 (39%)	18 (38%)	>0.99
Family history of sudden death†	5 (7%)	7 (15%)	0.23
Follow-up periods*	4.6±4.9	5.2±5.7	0.36
Symptoms after diagnosis†	12 (17%)	17 (35%)	0.03
Syncope	12	17	...
Aborted cardiac arrest	0	2§	...
Sudden cardiac death	0	2#	...

*The mean value±SD.

†Number of subjects and percentage in parenthesis.

‡Of 7 subjects with aborted cardiac arrest (ACA), 4 experienced both syncope and ACA.

#Of each 2 subjects with ACA or sudden cardiac death (SCD), all 4 subjects experienced syncope and ACA or SCD.

Genetic Testing

Referral for genetic testing was based on the opinion of the chief medical doctors in the present study. Pediatric cardiologists in the present study recommended genetic testing based on the following criteria: (1) for a patient in whom they had established a strong clinical index of suspicion for LQTS based on examination of the patient's clinical history, family history, and expressed ECG phenotype or; (2) for an asymptomatic patient with QT prolongation in the absence of other clinical conditions that might prolong the QT interval, as detailed in the recent consensus recommendation report.[24]

Genomic DNA was isolated from blood after obtaining written informed consent. Genetic screening for all exons of *KCNQ1*, *KCNH2*, *SCN5A*, *KCNE1*, *KCNE2*, *KCNJ2*, and *CAV3* was reperformed for the present study using polymerase chain reaction and direct DNA sequencing. When a patient was suspected to have Timothy syndrome, which is a multisystem disorder characterized by cardiac (QT prolongation and sometimes congenital heart diseases), hand/foot, facial, and neurodevelopmental features, the exons of *CACNA1C* were amplified. When a patient had a prolonged QT interval and hyperaldosteronism, the exon of *KCNJ5* was amplified. The exons of *ANKB*, *SCN4B*, *AKAP9*, and *SNTA1* were not analyzed because of a lack of reported cases of these mutations in the Japanese population. Genomic DNA was isolated using a QIAamp DNA Blood Midi Kit (Qiagen, Gaithersburg, MD). Polymerase chain reaction products were purified using AMPure (Beckman Coulter, Brea, CA). After treating with BigDye Terminator version 1.1 Cycle Sequence Kit (ABI, Warrington, United Kingdom) and BigDye X Terminator, direct sequencing was performed by a genetic analyzer, ABI3130x1 Genetic Analyzer (ABI). The study was approved by the Ethics Committee of the Kagoshima University Hospital between November 1993 and March 2005 and the National Hospital Organization Kagoshima Medical Center from April 2005.

Nucleotide changes reported as single nucleotide polymorphisms[18,25] were excluded from mutation analysis in the present study. However, amino acid changes of G643S in *KCNQ1*[26] and D85N in *KCNE1*[27] were included in the present study because previous reports have shown that these mutations are associated with an ≈30% reduction in potassium channel currents.[26,27] When multiple mutations were present, each mutation was counted in each genotype.

Mutations of High Probability of Pathogenicity

Mutations of a high probability of pathogenicity were based on data published by Giudicessi et al.[16] Radical mutations included splice-site, nonsense, frame-shift, and insertion/deletions.[16] Mutations of a high probability of pathogenicity in the present study were defined as those present in the subunit assembly domain of the C-terminal of *KCNQ1*, the Per-Arnt-Sim domain, Per-Arnt-Sim–associated C-terminal domain, and the cyclic nucleotide–binding domain of *KCNH2*. Mutations present in the transmembrane/linker/pore and C-terminal regions of *KCNQ1* and the transmembrane/linker/pore regions of *KCNH2* were also defined as those of a high probability of pathogenicity.[16] Remaining mutations were defined as those of uncertain significance.

Statistical Analysis

Differences in the mean values and prevalence values were examined using the Mann–Whitney *U* test and Fisher exact probability test, respectively. Tukey multiple comparison test was used to assess differences in the mean QTc values among first, seventh, and 10th graders. Statistical analysis was performed using IBM SPSS Statistics version 21.0 (IBM Japan, Ltd, Tokyo, Japan). A 2-tailed *P* value of <0.05 was considered statistically significant.

Results

Population

Characteristics of the 117 subjects, including 69 screened and 48 clinical patients, are shown in Table 1. Of the 48 subjects included in the clinical group, 36 were diagnosed with LQTS-related symptoms, 6 were diagnosed by familial study, and 6 were diagnosed by chance. Subjects who were diagnosed by chance included those who visited hospitals for medical checks and for examination of heart murmurs at 1 month (4 patients), those who had been followed with Kawasaki disease (1 patient), and as Ehlers–Donlos syndrome (1 patient). There were no differences in sex, mean QTc values, family history of LQTS, family history of sudden death, or follow-up period between the screened and clinical groups. The mean age was lower in the clinical group when compared with the screened group (P=0.04). Prevalence of subjects having LQTS-related symptoms before and after diagnosis was significantly lower in the screened group when compared with that in the clinical group (P<0.001 and P=0.03, respectively). Symptoms before and after diagnosis in the screened group were all syncope. Of 117 subjects, 2 subjects in the clinical group died. A girl had a history of aborted cardiac arrest at 2 months, and died suddenly in her sleep at 5 years of age. An 11-year-old boy had frequent symptoms and died suddenly during class. Genetic analysis failed to show the presence of any of the mutations analyzed in this study. The treatment of subjects with symptoms during follow-up period is shown in Table I in the online-only Data Supplement.

Mutations Determined in the Present Study

The yield of genetic testing in the present study by QTc values using Bazett formula is shown in Table II in the Data Supplement. The data show that there was no difference in yield between subjects with a QTc<500 ms and those with a QTc≥500 ms in both screened and clinical groups in the present study. Of 50 subjects who were screened and whose mutations were identified, 29, 18, and 3 subjects were screened in the first, seventh, and 10th grade, respectively. Their QTc values using Bazett formula were 491±35, 503±43, and 500±49 ms, respectively. There were no differences in QTc values among the screened periods. Of 117 subjects, mutations were found in 50 of 69 (72%) screened and 23 of 48 (48%) clinical subjects (Table III in the Data Supplement). The prevalence of LQT1, LQT2, and LQT3 between the 2 groups was not different. LQTS-related mutations in the present study are summarized in Table IV in the Data Supplement.

Genetic Characteristics of Subjects

All mutations found in *KCNQ1* from 18 mutations in the screened and 9 mutations in the clinical groups were located in regions of a high probability of pathogenicity (Table 2; Figure 1A and 1B). In the screened group, 8 mutations were located in the transmembrane/linker/pore regions and 10 were present in the C-terminal regions (Figure 1A). Three mutations were radical and 1 mutation was present in the subunit assembly domain. About the association between locations of mutation and the presence or absence of LQTS-related symptoms, 14 (78%) of 18 mutations were associated with the presence of symptoms in probands and family members, including 4 (22%) with family history of sudden death in the screened group. Eight of 9 mutations in the clinical group were associated with the presence of symptoms, and the remaining mutation was found in a subject who was diagnosed by a familial study.

Among mutations yielded in *KCNH2*, 13 (87%) of 15 mutations in the screened group and 5 (83%) of 6 mutations in the clinical group were located in regions of a high probability of pathogenicity (Table 2; Figure 2A and 2B). In the screened group, 1 mutation was both radical and present in the cyclic nucleotide–binding domain. Another 5 mutations were radical, and 1 each was present in Per-Arnt-Sim and Per-Arnt-Sim–associated C-terminal regions. However, only 4 (31%) of 13 mutations were associated with the presence of LQTS-related symptoms in probands or family members in the screened group. In the clinical group, 5 (83%) of 6 mutations were associated with the presence of symptoms in probands and family members, and the remaining mutation was found in a subject by ECG screening during a medical check-up at 1 month.

Discussion

Mutations in subjects with LQTS who were diagnosed by school-based ECG screening programs were mostly of high possibility of pathogenicity, similar to clinical subjects. Clinical background, such as QTc values, family history of LQTS, or sudden death, and follow-up periods, was not different between the 2 groups. However, prevalence of symptoms before and after diagnosis in the screened group was significantly lower when compared with the clinical group.

Table 2. Number of Patients With Mutations at High Risk in Each Group

Genes	Mutations	Screened Group	Clinical Group	*P* Value
*KCNQ1**	Radical mutation†	4	1	>0.99
	High probability‡	14	8	0.59
	Variants of uncertain significance	0	0	>0.99
KCNH2†	Radical mutation† and high probability‡	1	1	0.53
	Radical mutation†	5	1	0.66
	High probability‡	7	4	0.73
	Variants of uncertain significance	2	1	>0.99

*Variants of uncertain significance include mutations other than radical or of high probability of pathogenicity.

†Radical mutations include splice-site, nonsense, frame-shift, and insertion/deletions.

‡Mutations of high probability include subunit assembly domain, transmembrane/linker/pore, and C-terminal regions of *KCNQ1*, and the Per-Arnt-Sim (PAS) domain, PAS-associated C-terminal domain, the cyclic nucleotide–binding domain, transmembrane/linker/pore region of *KCNH2*.

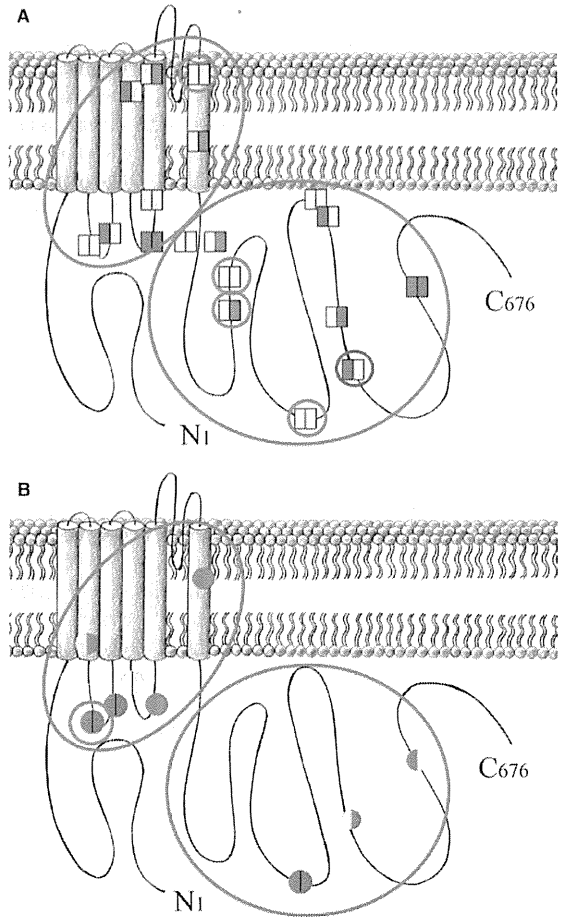

Figure 1. Topological depiction of *KCNQ1* in the present study in the screened (**A**) and clinical (**B**) groups. Mutations found in the screened group are shown as boxes (**A**) and those in the clinical group as circles (**B**). Each box or circle is divided into 2 parts: **left** and **right** sides. Each part represents the presence or absence of long-QT syndrome–related symptoms in probands (**left**) and family members (**right**), respectively. Green, brown, and red colors symbolize no symptoms, syncope or aborted cardiac arrest, and sudden death, respectively. Bold red circles surrounding mutations represent radical mutations. A bold blue circle represents subunit assembly domain. Two big purple circles symbolize locations of transmembrane/linker/pore and C-terminal regions of *KCNQ1*.

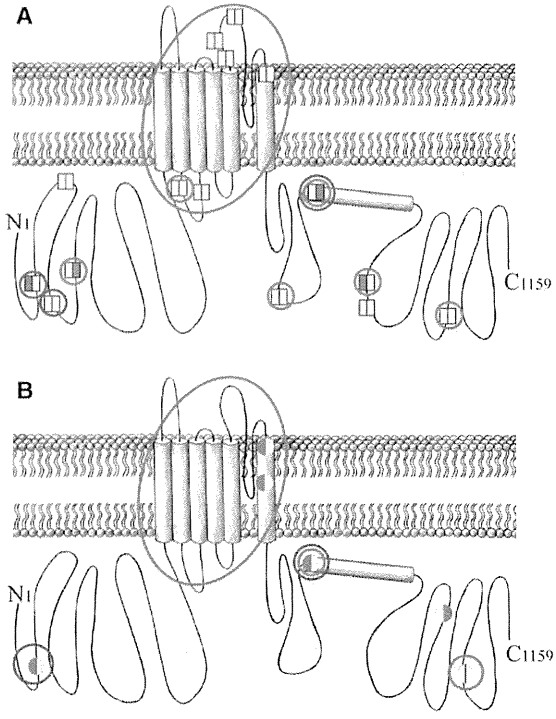

Figure 2. Topological depiction of *KCNH2* in the present study in the screened (**A**) and clinical (**B**) groups. Explanations of symbols and shapes are the same as in Figure 1. Bold blue circles surrounding mutations in this figure represent Per-Arnt-Sim (PAC), PAC-associated C-terminal, and cyclic nucleotide–binding domains, respectively, from the **left** side. A big purple circle symbolizes locations of transmembrane/linker/pore regions.

These data suggest that screening programs may be effective for early diagnosis of LQTS and prevention of symptoms, and that screened patients should be followed similar to clinical patients.

Clinical and genetic backgrounds of patients with LQTS have been reported widely for infants, children, adolescents, and adults.[5–9] These data were mostly based on symptomatic probands and family members. Few data are available on the genetic background of subjects who were diagnosed by ECG screening programs. Schwartz et al[12] reported that LQTS-related mutations were identified in 16 neonates of 43 080 who underwent neonatal ECG screening; 8 *KCNQ1*, 5 *KCNH2*, and 1 each of *KCNE1* and *KCNE2*. One infant had a digenic mutation of *KCNQ1* and *KCNH2*.

A school-based ECG screening program for heart diseases was initiated in 1994 for first, seventh, and 10th graders in Japan. The program screened subjects with QT prolongation. However, few studies have confirmed the genetic background in these screened subjects.[13,14] Hayashi et al[13] reported that mutations were identified in 3 subjects with high or intermediate probabilities of LQTS using Schwartz criteria from 7961 school children; all 3 mutations were present in *KCNH2*. Yasuda et al[14] reported that *KCNQ1* mutations were found in 8 of 13 pediatric patients and that 7 of 8 patients were diagnosed by the ECG screening program.

In the present study, a relatively large number of subjects, who were diagnosed by ECG screening programs accompanied by genetic testing, were included. The clinical backgrounds of the screened subjects, such as QTc values, family history of LQTS, or sudden death, were similar to clinical subjects. All 16 mutations in the *KCNQ1* gene in the screened group were radical or of high probability of pathogenicity similar to the clinical group. The ratio of mutations of radical and of high probability of pathogenicity in the *KCNH2* gene in the screened group (13/15; 87%) was remarkably similar to that in the clinical group (5/6; 83%). These data suggest that pediatricians, who asked for genetic testing in the present study, chose patients with similar clinical backgrounds in both groups, and that demand for genetic testing was more

prevalent in screened patients when compared with clinical patients when ECG screening was developed in Japan.

Conversely, prevalence of symptoms before and after diagnosis was significantly lower in the screened group when compared with that in the clinical group. A low prevalence of symptoms before diagnosis suggests that the ECG screening program is effective for early diagnosis of LQTS. The reason for low prevalence of symptoms after diagnosis in the screened group is uncertain. Doctors may recommend pediatric patients with LQTS and their parents adopt changes to their lifestyles, for example, not doing vigorous exercise, not swimming a lap, and not diving,[28] in both the screened and clinical subjects. The precise reason remains to be clarified.

The reason for no difference in the prevalence of family history between the screened and clinical groups is unclear. The authors posit that even now in Japan the general population may not be familiar with LQTS, and that the parents in the present study did not think that syncope in their children was a serious condition. In addition, they may have been unaware that LQTS is an inherited disease. The reason of the high prevalence of family history of LQTS in the screened group is also unclear. The authors speculate that doctors did not ask the parents (grandparents of the probands in the present study) to perform familial studies 2 or 3 decades ago, when parents of the probands of the present study and their family members experienced symptoms at younger ages; however, no data were obtained addressing this hypothesis from the families.

There are some limitations of the current study. First, we did not discuss subjects with the *SCN5A* gene. One fourth of pediatric patients with LQTS had the *SCN5A* gene. We need similar algorithms designed to guide the interpretation of genetic testing results for the *SCN5A* mutation and to determine the possibility of pathogenesis in patients with SCN5A in the future. Second, the clinical group showed a low rate (48%) of genotypic determination. We could not find mutations in 2 cases of death in the present study. The reasons for this are unclear. One potential reason was that we did not screen copy number variations in genes associated with LQTS.[29,30] Eddy et al[29] and Barc et al[30] reported that 3 of 26 (12%) and 3 of 93 (3%) unrelated mutation-negative probands showed copy number variations, indicating that some mutation-negative patients may have copy number variations. Another reason may be that numerous previously undetected mutations exist in symptomatic patients.

In conclusion, mutations in subjects with LQTS who were diagnosed by screening programs had a high probability of pathogenicity similar to clinical subjects. Clinical backgrounds were not different although the prevalence of symptoms before and after diagnosis in the screened group was significantly lower when compared with that in the clinical group. These data suggest that the school-based screening program may be effective for early diagnosis of LQTS and prevention of symptoms, and that screened patients should have follow-up equivalent to clinical patients.

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Disclosures

None.

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CLINICAL PERSPECTIVE

This study aimed to determine the genetic characteristics of 69 pediatric patients with long-QT syndrome who were diagnosed by a school-based screening program (screened group) and in whom genetic testing was performed. The screened group was compared with 48 subjects who visited hospitals because of the presence of long-QT syndrome–related symptoms, familial history, or who were diagnosed by chance (clinical group). A recently developed algorithm, designed to guide the interpretation of genetic testing results for *KCNQ1* and *KCNH2*, enabled us to classify the mutations as probably pathogenic or variant of uncertain significance. Using the algorithm, the authors found that of mutations yielded in *KCNQ1* or *KCNH2*, 31 of 33 (94%) mutations in the screened group and 15 of 16 (94%) mutations in the clinical group were radical and of high probability of pathogenicity. They also found that prevalence of symptoms before ($P<0.0001$) and after ($P=0.03$) diagnosis was significantly lower in the screened group when compared with that in the clinical group although the QTc values, family history of long-QT syndrome, sudden death, and follow-up periods were not different between the groups. Demand for genetic testing is now more prevalent in screened patients when compared with clinical patients because ECG screening was developed in Japan. This study may help to clarify the benefits of ECG screening. In addition, this study provides valuable genetic information and confirms that patients identified by ECG screening have the condition and are similar in many ways to those identified via a clinical setting.

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High Prevalence of the *SCN5A* E1784K Mutation in School Children With Long QT Syndrome Living on the Okinawa Islands

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Background: Genetic testing for long QT syndrome (LQTS) is now in clinical practice. We conducted molecular genetic analyses to definitively diagnose LQTS and to determine its subtypes for gene-specific treatment. We conducted a retrospective study to determine the characteristics of schoolchildren with LQTS living on the Okinawa Islands.

Methods and Results: The study population included children identified in a school-based electrocardiographic (ECG) screening program for cardiovascular diseases who were referred to Okinawa Children's Medical Center between 2007 and 2012; 23 children met the diagnostic criteria for LQTS. Of them, 17 were genotype-positive and 14 were found to harbor the *SCN5A* E1784K mutation exclusively among the LQTS genotype-positive children. The children were divided into genotype-positive and -negative groups. Clinical characteristics and ECG data were analyzed and compared. The median Schwartz score was 3. The median QT interval was 521 ms.

Conclusions: The major finding is that the prevalent subtype of LQTS in Okinawa is discordant with other cohorts living in other regions of Japan or overseas. We cannot exclude the possibility of the presence of a specific founder mutation in this geographically clustered population, particularly considering that the hospital is the only tertiary heart center for children in Okinawa. However, this uniquely high prevalence of the *SCN5A* E1784K mutation serves as a compelling justification to conduct a larger study.

Key Words: Arrhythmia; Long QT syndrome; Okinawa; *SCN5A*

The long QT syndrome (LQTS) is an inherited disorder of cardiac repolarization characterized by electrocardiographic (ECG) abnormalities, syncopal attacks, and risk of sudden death from ventricular tachyarrhythmias such as torsade de pointes.[1,2] Mutations in genes encoding cardiac ion channels and membrane adaptor proteins cause this syndrome. There are 13 distinct LQTS-susceptibility genes and mutations in the 3 most common LQTS subtypes (*KCNQ1*-mediated LQT1, *KCNH2*-mediated LQT2, and *SCN5A*-mediated LQT3) account for approximately 80% of clinically confirmed cases of LQTS.[3–5]

Editorial p????

Genotyping is used for adjunctive diagnosis as well as for gene-specific risk stratification and gene-specific therapy, particularly for LQTS.[5–8] We report here the overrepresentation of the LQT3 mutation detected by genetic testing conducted at a regional tertiary children's medical center on the Okinawa Islands of Japan, and describe the children's clinical characteristics and outcomes.

Methods

Patient Recruitment

Okinawa Children's Medical Center is the only tertiary hospital for children on the Okinawa Islands of Japan. It serves 1.4 million people and is a regional referral center that offers a comprehensive clinical practice of pediatric cardiology. A school-based ECG screening program for cardiovascular diseases aimed at schoolchildren in the 1st, 7th, and 10th grades is conducted all over Japan to identify heart problems. Schoolchildren are also checked for clinical and familial findings associated with LQTS, such as syncope, congenital deafness, and unexplained sudden cardiac death among family members. Children with positive findings are directed to referral

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Table 1. Demographic Data of Schoolchildren With LQTS on the Okinawa Islands

	Total	Gene positive	Gene negative	P value
n	23	17	6	
Age at 1 st referral (years)	9.7±3.4	9.4±3.6	10.8±3.4	NS
Sex (M/F)	15/8	11/6	4/2	NS
Follow-up duration, months	37±23	43±23	23±14	NS

Data given as mean±SD. LQTS, long QT syndrome.

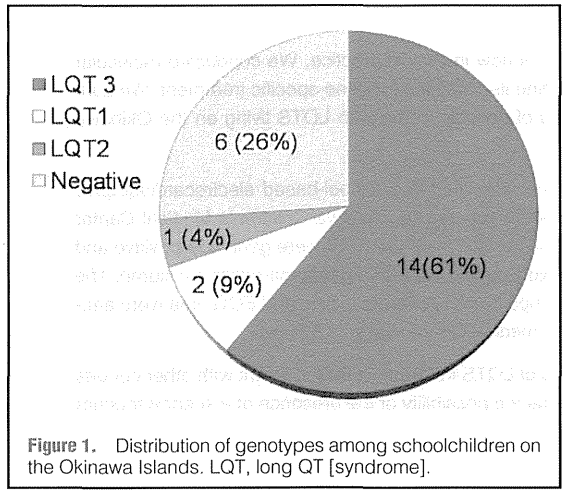

Figure 1. Distribution of genotypes among schoolchildren on the Okinawa Islands. LQT, long QT [syndrome].

hospitals.[9,10]

The present study population was retrospectively recruited from children who participated in the school-based ECG screening program and were referred to Okinawa Children's Medical Center between April 2007 and March 2013. The diagnosis of LQTS and screening for prolonged QT intervals was based on the judgment of the physician in each hospital that participated in the program in each area. For their final diagnosis, many physicians use a scoring system published in 1993.[11] Included were children who met the diagnostic criteria for LQTS ("Schwartz and Moss" score ≥3). This approach is similar to that described in a published study,[5] although the most recent HRS/EHRA/APHRS expert consensus statement recommends that the score is ≥3.5.[12]

Retrospective review was approved by the Institutional Review Board. The patients' characteristics that were reviewed included sex, age at diagnosis, presence or absence of syncope, seizures, aborted cardiac arrest, family history, and 12-lead ECG. We assigned a cumulative LQTS diagnostic score, which is derived in part from the corrected QT interval (QTc), symptoms, and family history.[11] The children were divided into genotype-positive and -negative subgroups (G-pos and G-neg) according to the results of genetic analysis. Clinical characteristics and ECG data were analyzed and compared between the subgroups.

Clinical Testing

Each patient referred to the center for the evaluation of LQTS had their history taken and underwent a physical examination and a supine ECG. The QT interval was measured with a standard 12-lead ECG using lead V5. The ECG measurements were conducted by K. T. who was blinded to each subject's genetic status before making the measurements. QT interval was measured manually and defined as the time interval between QRS onset and the point at which the isoelectric baseline intersected a tangential line drawn at the maximal downslope of the positive T wave. QT interval measurements represented the mean of 3 consecutive beats in lead V5. The QT interval was corrected for rate using the Bazett formula ($QT/\sqrt{RR}$, interval measured in seconds). The LQTS score was determined according to the LQTS diagnostic criteria described earlier. Diagnostic criteria included the QTc interval (450 ms), at least 1 symptomatic episode (bradycardia), or a family history of LQTS. A cumulative score ≥3 suggested an intermediate probability of LQTS.

Genetic Testing

Genetic testing was recommended according to the opinion of the chief physician who participated in the present study, and it was performed when the phenotype suggested LQTS according to the Schwartz score and clinical presentation or as detailed in the recent consensus report.[12] After written informed consent was given, genomic DNA was isolated from blood for direct sequencing of the complete *KCNQ1, KCNH2* and *SCN5A* genes in the National Cerebral and Cardiovascular Center, Osaka, Japan.

Statistical Analysis

The values of the continuous variables are presented as the mean±standard deviation or median and interquartile range. Categorical variables are represented by frequencies and percentages. Data were compared between the 2 groups using the following tests: t-test for normally distributed continuous variables, Mann-Whitney test for non-normally distributed data, and the chi-square or Fisher's exact test to compare the frequency distribution of categorical variables. Two-tailed $P<0.05$ was considered statistically significant. Analyses were performed using StatView J-5.0 PPC (SAS Institute Inc, Cary, NC, USA).

Results

Study Population

The study population comprised 23 children who met the criteria during the study period. Demographic data at referral are shown in Table 1.

A total of 17 (74%) children were positive in genetic testing (G-pos subgroup; 6 females) and 6 were negative (G-neg subgroup, 2 females). Mean age at referral was 9.7±3.4 years. Total follow-up period for the G-pos and G-neg groups was 43±23 months and 23±14 months, respectively. Figure 1 shows the distribution of genotypes. The G-pos subgroup comprised LQT type 1 (LQT1, n=2), LQT type 2 (LQT2,

Table 2. ECG Data and Symptoms at Referral of Schoolchildren With LQTS on the Okinawa Islands

	Total	Gene positive	Gene negative	P value
ECG findings				
Average QTc (ms)	510±44	516±45	495±39	NS
% w/QTc >480 ms	15 (65%)	12 (71%)	3 (50%)	NS
% w/Schwartz score ≥4	8 (35%)	6 (35%)	2 (33%)	NS
Late-appearing T wave	15 (65%)	14 (82%)	1 (17%)	<0.01
Symptoms				
Syncope, n	1	0	1	NS
Bradycardia, n	1	1	0	NS
Epilepsy	1	0	1	NS
Family history				
Positive for LQTS, n	8 (35%)	7 (41%)	1 (17%)	NS

ECG, electrocardiographic; LQTS, long QT syndrome; QTc, corrected QT interval.

Table 3. Symptoms Newly Developed in Schoolchildren With LQTS on the Okinawa Islands During Follow-up Period and Treatment

	Total	Gene positive	Gene negative	P value
Symptoms				
Syncope/presyncope, n (%)	3 (13)	2 (12)	1 (17)	NS
Bradycardia, n	4	2	2	NS
Epilepsy, n	2	1	1	NS
Treatment				
Medications, n (%)	9 (39)	7 (41)	2 (33)	NS
Mex, n	8	7	1	
BB, n	3	3	0	
Antiepileptic, n	2	1	1	
Pacemaker, n	1	1	0	NS

BB, β-blocker; LQTS, long QT syndrome; Mex, mexiletine.

n=1), and LQT type 3 (LQT3, n=14). LQT3 was the most prevalent subtype (82%) in the G-pos subgroup. Further, all patients with LQT3 were found to share the same *SCN5A* c. 5350G>A p.E1784K mutation within the sequence that encodes the transmembrane region of the SCN5A sodium ion channel.

ECG Data and Clinical Characteristics at Referral

Clinical and ECG data of the subgroups were analyzed, and the comparisons are shown in Table 2. The average QTc interval was 510 ms (516 ms vs. 495 ms, NS). The frequency of patients with QTc >480 ms and with a Schwarz and Moss score >4 was not significantly different between the groups.

Although the findings were somewhat subjective for QT morphology, late-appearing T wave was more frequent in the G-pos subgroup (82% vs. 17%, P<0.01). There was no Brugada syndrome (BrS)-type ECG in the cohort at referral. No provocative test with sodium-channel blockers was performed. Each child was asymptomatic at presentation except for 1 in the G-neg subgroup; 1 child in the G-pos subgroup exhibited sinus bradycardia, and 1 child in the G-neg subgroup was epileptic. All but 2 children were unrelated. To the best of our knowledge, all parents of the probands were descendants of the Okinawa Islands. There was no identifiable common relative for this cohort after pedigree construction. A familial history of LQT was identified in 35% of the children, and its presence trended toward an increase among the G-pos children compared with the G-neg children (41% vs. 17%). Pacemakers were implanted in selected children based on screening of their first-degree relatives (unpublished data).

Clinical Outcome and Treatment During Follow-up

During the study period, no child died from sudden cardiac arrest, although syncope or presyncope occurred (n=2 vs. n=1, NS) (Table 3). One child in each subgroup developed epilepsy, which was diagnosed using electroencephalography. Treatment with an antiepileptic drug controlled their symptoms, and 1child was the subject of another report.[13] Two children experienced bradycardia on Holter monitoring, and 1 child in the G-pos group required pacemaker therapy for symptomatic sick sinus syndrome. The detailed clinical features of patients with newly developed symptoms during the follow-up period are presented in Table 4.

Treatment Nine (39%) of 23 patients received pharmacotherapy (7 and 2 in the G-pos and G-neg subgroups, respectively). The indication for pharmacotherapy in asymptomatic LQTS patients considered at high risk is QTc >500 ms, if the patient also agrees to keep taking the medication. One patient in each group received an antiepileptic agent. Mexiletine was administered to 7 patients in the G-pos subgroup (with β-blockers for 3) and 1 in the G-neg subgroup without β-blocker.

In the G-pos subgroup, QTc significantly decreased (from 553 ms to 491 ms; P=0.02) in the 7 patients receiving mexi-

Table 4. Characteristics of Newly Developed Symptoms in Schoolchildren With LQTS on the Okinawa Islands During Follow-up Period

Subtype	Age (years)	Sex	Gene test	QTc at referral (ms)	QTc at follow-up (ms)	Symptoms	Cardiovascular treatment
G-pos	13	F	KCNQ1	531	524	Resuscitation drowning	Mex, BB
G-pos	17	F	E1784K	591	502	Syncope VT	Mex, BB, pacemaker
G-pos	11	F	E1784K	565	423	Epilepsy	Mex
G-neg	12	M	Negative	556	553	Syncope with exertion	Mex
G-neg	10	M	Negative	492	505	Epilepsy	No

VT, ventricular arrhythmia. Other abbreviations as in Tables 2,3.

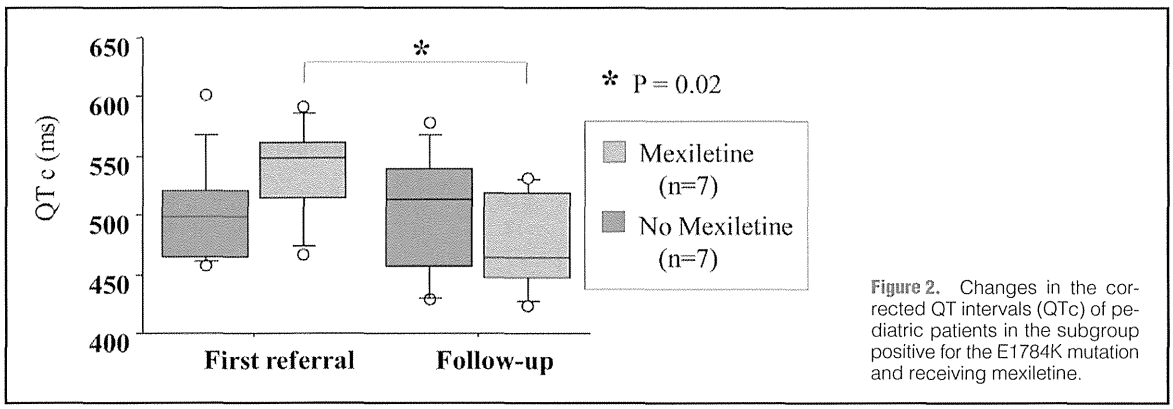

Figure 2. Changes in the corrected QT intervals (QTc) of pediatric patients in the subgroup positive for the E1784K mutation and receiving mexiletine.

letine, whereas the change in QTc (from 497 ms to 508 ms) was not significant in the 7 patients receiving no medication (Figure 2). All but 1 child on medication in the G-pos group showed no BrS-type ECG on the third-right precordial leads.

Discussion

The most significant finding of this retrospective study of a small cohort is that the proportion of patients with LQT3 with the *SCN5A* mutation was significantly higher than previously reported for patients living in other regions of Japan or in other countries. Further, the course of LQTS probands with the E1784K mutation was relatively benign during the study period.

Prevalence of LQT3

The children referred to us were screened according to the judgment of the physicians who participated in the program in each area. Therefore, the exact number of the entire population of participants in this study is unknown. According to another study, the prevalence of long QT was 1 in 1,164 among 7th graders aged 12 years.[14]

In general, approximately 90% of patients with genotyped LQTS carry a mutation in *KCNQ1*, *KCNH2* (*HERG*), or *SCN5A*, which is responsible for the LQTS subtype.[5,8] The most prevalent form of LQTS appears to be caused by mutations in *KCNQ1*, which encodes the potassium channel (LQT1).[7,9,10,15] Mutations in *SCN5A* (LQT3) are responsible for only a small proportion (10–20%) of genotyped patients with LQTS.[16,17] In sharp contrast to these previous findings, the present study describes a predominance of LQT3 (82% of genotyped patients with LQTS) among schoolchildren living on the Okinawa Islands. Even more striking is the exclusive presence of the *SCN5A* E1784K mutation that causes LQT3. Yoshinaga et al[9] recently reported the genetic characteristics of children and adolescents with LQTS diagnosed by school-based ECG screening programs. The incidence of LQT3 was 23% of all subjects in that study population with LQTS. An E1784K mutation was detected only in 1 patient. Blaufox et al published a multicenter study of patients residing in North America that showed that an E1784K mutation is present in 13 (30%) of 43 pediatric patients with LQT3.[18]

In patients with the *SCN5A* E1784K mutation, overlap of sinus node dysfunction and BrS is relatively common.[19] Regarding the prevalence of LQT3 in other Asian countries such as Taiwan, China, and South Korea, we speculate that the high prevalence of LQT3 is similar to that of BrS in Asia.[20] To the best of our knowledge, only a limited number of reports have addressed the prevalence of LQT3 in Asian countries. For example, the *SCN5A* mutation was detected in 17.5% of all genotyped patients with LQTS residing in South Korea.[21] The mutation rate of *KCNQ1* (6.4%) and *KCNH2* (6.4%) was lower in the Chinese population compared with those of North America or Europe.[22] The true prevalence of LQT3 in Asia is unknown, but we believe that it is reasonable to conclude that LQT3 may be prevalent in China.

The high prevalence of LQT3 in Okinawa may be accounted for, in part, by the potential bias introduced by studying a small number of patients at a single center. A second possible cause is a founder mutation, which is defined as disproportion-

ate genetic representation resulting from an affected common ancestor who established a new population, in contrast to the presence of the same mutation among individuals. Such a regional predominance in frequency is associated with LQT1.[23–25] There is a report that describes a large Dutch family with a founder mutation that uniquely overlaps those of LQT3, BrS, and progressive cardiac conduction defects.[26]

The subtropical Okinawa Islands are located 2,000 km south of mainland Japan between the East China Sea and the Pacific Ocean. Although haplotype analysis was not performed, a common ethnic heritage for the inhabitants of the Okinawa Islands is consistent with a founder effect. The study subjects described here are unrelated, except for 2. Therefore, the predominant genotype of the children with LQTS studied here strongly suggests community clustering that may represent a founder effect of the same *SCN5A* mutation.

Clinical Characteristics of Patients and Their Treatment

In the present study, most patients were asymptomatic at initial diagnosis, and the disease may have been relatively benign at this time. However, 2 patients experienced bradycardia during the follow-up period, and 1 required implantation of a pacemaker. Zareba et al reported that a cardiac event will occur after adolescence in patients with LQT3.[27] The symptoms of patients with LQT3 generally appear after puberty, and in BrS, the electrophysiological phenotype is most prevalent in the 3rd decade. The developmental characteristics of LQT3 and BrS show age-dependent penetrance.[28] The late presentation and frequent asymptomatic status of carriers is consistent with a benign disease with possible post-reproductive pathogenicity in the population of the Okinawa Islands.

To date, 8 of 15 patients in the present study received mexiletine. Only 1 patient exhibited a BrS-type ECG while taking this medication. Bradycardia was detected using the third right-precordial lead in the single patient with a BrS-type ECG. Patients with LQT3 without cardiac events in the first year of life respond extremely well to therapy with β-blockers, left cervical sympathetic denervation, or both.[29] Mexiletine is considered the medication of choice for treating LQT3.[30,31] However, 2 studies recommend caution when treating LQT3 because a patient's response depends on the type of mutation.[32,33] Further, an in-vivo study showed that patients with the *SCN5A* E1784K mutation respond to mexiletine.[19] The diagnosis of LQT3 does not necessarily indicate treatment with an implantable cardioverter-defibrillator. For this reason, Schwartz et al recommend a staged strategy.[29]

Study Limitations

This was a small and retrospective cohort study. Although the study groups should include patients with LQT3 as well as those positive and negative for mutation, the distribution of the prevalence of genotype-positive subtypes was unpredictably imbalanced and unique. The prevalence of LQT3 (exclusively the E1784K mutation) was predominantly high in genotype-positive patients; therefore, we conclude that our classification was justified. Moreover, despite the possibility of the enrollment of patients with a specific founder mutation and considering that the hospital is the only tertiary heart center for children in Okinawa, our findings of a unique and highly prevalent *SCN5A* mutation (E1784K) serve as a compelling justification to conduct larger or long-term studies.

Conclusions

We were unable to determine why the prevalence of the *SCN5A* E1784K mutation was uniquely high among schoolchildren living on the Okinawa Islands. The prognosis of these patients is relatively benign; however, the prognosis for adults is unknown. Because of the geography of Okinawa, its population should be ideal for studies on gene modifiers with a focus on the prevalent *SCN5A* mutation.

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