JACC: CLINICAL ELECTROPHYSIOLOGY VOL. 4, NO. 4, 2018

PUBLISHED BY ELSEVIER

Clinical Outcomes and Modes of Death in Timothy Syndrome

A Multicenter International Study of a Rare Disorder

Keith A. Dufendach, MD,[a,b] Katherine Timothy, BS,[c] Michael J. Ackerman, MD, PhD,[d,e,f] Benjamin Blevins, MD,[g] Andreas Pflaumer, MD,[h] Susan Etheridge, MD,[c] James Perry, MD,[i] Nico A. Blom, MD,[j,k] Joel Temple, MD,[l] Devyani Chowdhury, MD,[m] Jonathan R. Skinner, MBChB, DCH, MRCP,[n] Christopher Johnsrude, MD,[o] Andras Bratincsak, MD,[p] J. Martijn Bos, MD, PhD,[d,e,f] Maully Shah, MBBS[b]

ABSTRACT

OBJECTIVES The objective of this study was to evaluate contemporary clinical outcomes and identify triggers for arrhythmias or sudden death in an international cohort of Timothy Syndrome (TS) patients including those with novel TS-associated CACNA1C mutations.

BACKGROUND TS is an extremely rare genetic disorder of the L-type cardiac channel $Ca_v1.2$ encoded by *CACNA1C*. The syndrome is characterized by multisystem abnormalities consisting of QT prolongation, congenital heart defects, syndactyly, facial dysmorphism, and neurological symptoms.

METHODS Patients diagnosed with TS between January 1, 1994, and April 1, 2016, from 12 international tertiary care pediatric centers were included in this retrospective study. Data were gathered via survey from the patients' electrophysiologists.

RESULTS Seventeen patients diagnosed with TS were identified. Length of follow-up was 4.9 years (range 3.0 to 19.0 years). Mean QTc was 640 ms (range 500 to 976 ms). All patients were treated with beta-blockers; 13 patients (76%) were also treated with an implantable defibrillator. Eleven patients experienced an episode of aborted cardiac arrest, 6 associated with general anesthesia and 2 with hypoglycemia. Four patients died suddenly due to ventricular fibrillation, 2 of whom had associated hypoglycemia.

CONCLUSIONS This study shows that mortality in TS patients is due to multifactorial mechanisms, which include ventricular arrhythmias, pulseless electrical activity, and hypoglycemia. A simple nomenclature for ongoing studies of TS and related syndromes is described. A worldwide prospective registry is needed for continued exploration of this syndrome. (J Am Coll Cardiol EP 2018;4:459–66) © 2018 by the American College of Cardiology Foundation.

From the [a]Department of Pediatrics, Division of Pediatric Cardiology, Perelman School of Medicine at the University of Pennsylvania, Philadelphia, Pennsylvania; [b]Department of Pediatrics, Division of Pediatric Cardiology, Children's Hospital of Philadelphia, Philadelphia, Pennsylvania; [c]Department of Pediatrics, Division of Pediatric Cardiology, University of Utah, Salt Lake City, Utah; [d]Department of Cardiovascular Diseases, Division of Heart Rhythm Services, Mayo Clinic, Rochester, Minnesota; [e]Department of Pediatrics, Division of Pediatric Cardiology, Mayo Clinic, Rochester, Minnesota; [f]Department of Molecular Pharmacology & Experimental Therapeutics, Windland Smith Rice Sudden Death Genomics Laboratory, Mayo Clinic, Rochester, Minnesota; [g]Department of Pediatrics, Division of Pediatric Cardiology, Naval Medical Center San Diego, San Diego, California; [h]Department of Pediatrics, Division of Pediatric Cardiology, Royal Children's Hospital, MCRI, and University of Melbourne, Melbourne, Australia; [i]Department of Pediatrics, Division of Pediatric Cardiology, Rady Children's Hospital/UC San Diego, San Diego, California; [j]Department of Medicine, Division of Pediatric Cardiology, Leiden University Medical Center, Leiden, the Netherlands; [k]Department of Medicine, Division of Pediatric Cardiology, Academic Medical Center, Amsterdam, the Netherlands; [l]Nemours/Alfred I. duPont Hospital for Children, Wilmington, Delaware; [m]Cardiology Care for Children, Lancaster, Pennsylvania; [n]Department of Pediatrics, Division of Pediatric Cardiology, Starship Children's Hospital, Auckland, New Zealand; [o]University of Louisville, Louisville, Kentucky; and the [p]Pediatric and Adult Congenital Cardiology, Kapi'olani Medical Specialists, Honolulu, Hawaii. This study was approved by the Pediatric and Congenital Electrophysiology Society (PACES). The views expressed in this article are those of the authors and do not reflect the official policy or position of the Department of the Navy, Department of Defense, or the United States Government. Dr. Ackerman has received consultant fees from Boston Scientific, Gilead Sciences, Medtronic, and St. Jude Medical; and royalties from Transgenomic. Dr. Shah has received a research grant unrelated to the study

ISSN 2405-500X/$36.00 http://dx.doi.org/10.1016/j.jacep.2017.08.007

JACC: CLINICAL ELECTROPHYSIOLOGY VOL. 4, NO. 4, 2018
APRIL 2018:459–66

ABBREVIATIONS AND ACRONYMS

ACA = aborted cardiac arrest

AED = automatic external defibrillator

AV = atrioventricular

CHD = congenital heart disease

CIED = cardiac implantable electronic device

ECG = electrocardiogram

GA = general anesthesia

ICD = implantable cardioverter-defibrillator

LCSD = left cervical sympathetic denervation

LQTS = long QT syndrome

SCD = sudden cardiac death

TdP = torsades de pointes

TS = Timothy syndrome

TWA = T-wave alternans

VF = ventricular fibrillation

VT = ventricular tachycardia

Timothy syndrome (TS) is an extremely rare genetic disorder characterized by myriad multisystem abnormalities, consisting of a cardiac phenotype that universally includes QT prolongation and potentially congenital heart disease (CHD) and/or cardiac hypertrophy, syndactyly, facial dysmorphism, and a neurological phenotype that can include autism, seizures, and intellectual disability. The original genetic discovery attributed the predominant cause to a recurrent de novo heterozygous missense mutation, p.Gly406Arg, in the alternatively spliced exon 8A of *CACNA1C* (1). Subsequently, 2 additional mutations were identified in exon 8 (p.Gly402Ser and p.Gly406Arg) associated with a slightly different phenotype than the original exon 8A-localizing missense mutation (2–4). With the advent of whole exome sequencing, other novel *CACNA1C* mutations have emerged, leading to an elucidation of phenotypic variants (5–11). Extreme QT prolongation is typical in TS and predisposes to ventricular fibrillation (VF), cardiac arrest (ACA), and sudden cardiac death (SCD). According to previous reports, most patients die before the third year of life despite medical treatment with beta-blockers (1). However, clinical outcome data reflecting current management strategies of this high-risk cohort have not been reported in the past decade.

We evaluated clinical outcomes and triggers for arrhythmia or sudden death in an international cohort of patients with TS, including those with novel TS-associated *CACNA1C* mutations. This case series provides a contemporary report of clinical course and outcomes in TS patients, along with updated management strategies and prognostic findings.

METHODS

STUDY COHORT. Patients diagnosed with TS from January 1, 1994, to April 1, 2016, from 12 international tertiary care pediatric centers were included in this retrospective study. De-identified data of TS patients from participating institutions were transferred electronically to the coordinating center and included baseline demographics, clinical characteristics, electrocardiogram (ECG) findings, genetic test results, treatment strategies, and outcomes. Additionally, hospitalization records including types of procedures and anesthetics were evaluated, along with the response and complications related to therapy.

Specific clinical characteristics evaluated included syndactyly, facial anomalies, abnormal dentition, baldness at birth, neurodevelopmental delay, CHD, suspected immune disorders, and hypoglycemia. The diagnosis of noncardiac manifestations, such as neurodevelopmental delay, suspected immune disorders, and hypoglycemia, were made based on the assessment by the patient's treating clinician (12,13).

INCLUSION CRITERIA. Patients were included in this study if they met the clinical criteria for TS with or without a confirmed genetic diagnosis as follows (1): QTc ≥480 ms and 1 or more of the following clinical features: 1) unilateral or bilateral cutaneous syndactyly; 2) typical TS facial and dental anomalies; 3) neurological symptoms including autism, seizures, intellectual disability, and hypotonia; and 4) CHD.

ECG AND RHYTHM ABNORMALITIES. The standard pediatric ECG was reviewed for measurements and rhythm analysis. QTc was calculated according to the Bazett formula on a standard pediatric ECG. Maximum QTc recorded was used for analysis in this study. The T waves were analyzed for the presence of T-wave alternans (TWA). Bradyarrhythmias were defined as: 1) pauses in the rhythm >3 s and 2) atrioventricular (AV) block. Tachyarrhythmias were classified as: 1) torsades de pointes (TdP); 2) ventricular tachycardia (VT); 3) VF; and 4) atrial tachycardia/atrial fibrillation. Rhythm abnormalities data were obtained from documentation in medical charts, implantable cardioverter-defibrillator (ICD) analysis, and automatic external defibrillator (AED) rhythm analysis.

GENETIC TESTING. Genetic testing approaches included: 1) targeted analysis for pathogenic variants of *CACNA1C*, followed by whole gene sequence analysis if no pathogenic variant is found; 2) sequence analysis of *CACNA1C* followed by gene-targeted deletion/duplication analysis if no pathogenic variant is found; 3) use of a multigene panel that includes *CACNA1C* and other genes of interest; and

presented in this manuscript from Medtronic, Inc. Dr. Temple has served as a consultant for St. Jude Medical. All other authors have reported that they have no relationships relevant to this paper to disclose.
All authors attest they are in compliance with human studies committees and animal welfare regulations of the authors' institutions and Food and Drug Administration guidelines, including patient consent where appropriate. For more information, visit the *JACC: Clinical Electrophysiology* author instructions page.

Manuscript received January 31, 2017; revised manuscript received August 21, 2017, accepted August 22, 2017.

4) whole exome sequencing. Mosaicism was defined if the mutation had only a limited presence in the derived DNA, suggesting that 2 genetically distinct populations of cells coexisted within an individual's somatic cells and/or gametes.

CARDIAC IMPLANTABLE ELECTRONIC DEVICES. For patients with ICDs and pacemakers, device interrogation reports were analyzed for the presence of ventricular arrhythmias. The cardiac implantable electronic devices (CIEDs) were analyzed for the presence of: 1) TdP; 2) VT; 3) VF; and 4) supraventricular arrhythmias. ICD therapies were analyzed to determine whether therapies were appropriate and defibrillation successfully terminated the documented ventricular arrhythmia. Implantation of ICDs was classified as for either primary or secondary prevention based on whether they were implanted before (primary prevention) or after (secondary prevention) an ACA or documentation of sustained VT.

STATISTICAL ANALYSIS. Data are expressed as mean ± SD, and median values are given when appropriate.

RESULTS

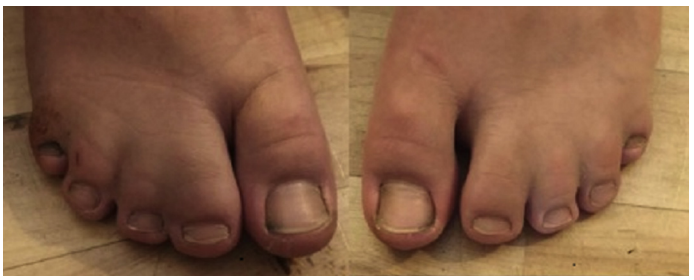
PATIENT CHARACTERISTICS. Seventeen patients (88% male) met study inclusion criteria. Age at diagnosis of TS was 17 ± 22 months (range birth to 76 months). The median follow-up period was 4.9 years (range 3.0 to 19.0 years), with a median age of 6.4 years at last follow-up.

Among all TS patients, a prenatal diagnosis of bradycardia secondary to functional 2:1 AV block was made in 4 patients (24%). Simple cutaneous syndactyly involving soft tissue only was present in 14 patients (82%) (Figure 1), neurodevelopmental delay in 11 (65%), facial anomalies in 8 (47%), abnormal dentition in 6 (35%) (Figure 2), baldness in 5 (29%), hypoglycemia in 7 (39%), CHD in 5 (29%), and suspected immunological disorder in 5 (29%). At last follow-up, 14 patients (77%) were alive. Patient characteristics are given in Table 1.

ECG AND RHYTHM CHARACTERISTICS. Mean QTc values on the resting ECG were 640 ± 108 ms (range 500 to 976 ms). TWA was present in 8 (47%), functional 2:1 AV block in 8 (47%), and bradycardia due to sinus pauses in 1 (6%), and was present at any time during the course of cardiac monitoring.

GENETIC TESTING. Details of patient mutations are summarized in Table 2. All 17 patients underwent genetic testing and had mutations in the *CACNA1C*-encoded cardiac calcium channel, although 1 was for research purposes and the results were not available.

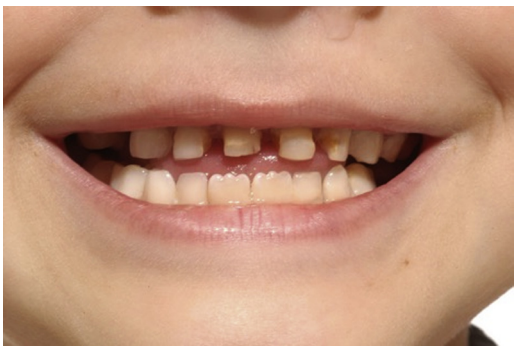
FIGURE 1 Cutaneous Syndactyly of the Feet in Patient #4



A missense mutation, p.Gly406Arg in the alternatively spliced exon 8 (exon 8A), was present in 10 patients (59%) (1–3). Three previously unreported mutations are detailed in this report. One mutation (p.Ser405Arg) in *CACNA1C* exon 8 was present in 2 related patients. Additionally, a p.Gly402Arg mutation in exon 8 was identified in 2 unrelated patients. Another novel mutation (p.Cys1021Arg) in exon 19 was identified in 1 patient with a classic TS phenotype of prolonged QT, TWA, and cutaneous syndactyly. In 2 children (12%), the TS-associated mutation was inherited from a parent with confirmed germline mosaicism. Of note, patient #12 had an additional mutation in *KCNQ1* that was inherited from an asymptomatic father but was not diagnosed with concomitant LQT1. Finally, 7 of the 11 patients with ACA/SCD had exon 8A/8-localizing *CACNA1C* mutations.

TREATMENT STRATEGIES. Antiarrhythmic medications. All patients were treated with beta-blockers once the diagnosis of TS was clinically established (Table 2). Propranolol was used in 9 patients (53%), atenolol in 4 (24%), and nadolol in 4 (24%). Eight patients (#1, #2, #3, #5, #6, #7, #12, and #16) were taking beta-blockers at the time of the cardiac event (ACA or SCD). Beta-blocker therapy for Patient #2 was omitted on the

FIGURE 2 Abnormal Dentition With Widely Spaced Teeth in the Upper Jaw in Patient #2



JACC: CLINICAL ELECTROPHYSIOLOGY VOL. 4, NO. 4, 2018
APRIL 2018:459–66

TABLE 1 Patient Characteristics

Patient #	Age at Diagnosis (months)	Max QTc (ms)	TWA	2:1 AV Block	CHD	Syndactyly	FA	NDD	Baldness	Immune Disorder	Hypoglycemia
1	6	690	No	No	PDA	Yes	No	No	No	No	No
2	8	710	Yes	Yes	PDA	Yes	Yes	Yes	Unknown	No	No
3	0	570	Yes	Yes	PDA	Yes	No	Unknown	No	Suspected	No
4	6	500	No	Yes	No	Yes	Yes	Yes	Unknown	Suspected	Yes
5	0	650	Yes	Yes	No	Yes	No	Unknown	No	No	Yes
6	8	550	Yes	No	No	Yes	Yes	Yes	Yes	Suspected	No
7	0	976	Yes	Yes	VSD	Yes	Yes	Yes	Yes	No	No
8	76	566	No	No	No	Yes	No	Yes	Unknown	No	No
9	11	640	Yes	Yes	PFO	Yes	No	No	Yes	Suspected	Yes
10	0	711	No	Yes	No	No	Yes	Yes	Yes	Suspected	Yes
11	34	606	Yes	No	No	Yes	No	No	No	Unknown	No
12	53	560	No	No	No	Yes	No	Yes	No	No	No
13	41	665	Yes	No	No	No	No	Yes	No	No	No
14	12	744	No	Yes	No	Yes	Yes	Yes	Unknown	No	Yes
15	0	546	No	Yes	No	No	No	No	No	No	No
16	6	600	No	No	No	Yes	Yes	Yes	Unknown	Unknown	Yes
17	24	590	Yes	No	No	Yes	Yes	Yes	Yes	Unknown	Yes

AV = atrioventricular; CHD = congenital heart disease; FA = facial abnormalities; NDD = neurodevelopmental delay; PDA = patent ductus arteriosus; PFO = patent foramen ovale; TWA = T-wave alternans; VSD = ventricular septal defect.

day of his surgical procedure, and he had an ACA while under GA. Four patients (#1, #9, #10, and #13) presented with ACA and were diagnosed with TS after the event and therefore were not taking beta-blockers at the time of their cardiac event. These 5 patients were started on beta-blockers after the diagnosis of TS was established, and as noted earlier Patient #1 had another cardiac arrest while taking beta-blockers. Three patients were also treated with mexiletine. One patient had recurrent arrhythmia during a trial off mexiletine, so the medication was restarted.

ICD insertion. A total of 13 patients (76%) had an ICD. ICD insertion was performed for primary prevention in 8 (61%) and secondary prevention in 5 (38%). ICD insertions were via a sternotomy in 6, with the high-voltage coil placed in the posterior pericardium and an abdominal ICD generator. Two patients had an ICD system that included an abdominal generator and a subcutaneous high-voltage coil, 4 had epicardial defibrillation patches with an abdominal generator, and 1 had a transvenous endocardial ICD system. No ICD complications were reported. Four patients (#5, #7, #13, and #16) had appropriate ICD discharges (3 VF, 1 TdP), which resulted in successful defibrillation in 2 patients (#7 and #13) and refractory VF leading to death in the other 2. The appropriate ICD discharge rate was 0.06/yr. No inappropriate ICD discharges were reported in the data. There were no ICD-related infections.

Pacemakers. Four patients (#3, #5, #10, and #15) had a permanent pacemaker inserted for functional 2:1 AV block, 2 of whom (#5 and #10) went on to have an ICD inserted (1 for primary prevention and 1 for secondary prevention of SCD). There were no pacemaker-related infections.

Left cervical sympathetic denervation. Three patients (#3, #7, and #14) underwent left cervical sympathetic denervation (LCSD). The latter 2 patients also had an ICD inserted at the time of LCSD.

TACHYARRHYTHMIAS, ACA, AND SCD. A total of 11 patients (65%) had ACA or SCD. Rhythm documentation was available in 10 patients (Table 3). One patient (#1) also had documented atrial fibrillation. The potential proarrhythmic triggers for the documented ventricular tachyarrhythmias were general anesthesia (GA) in 6, hypoglycemia in 2, and exertion in 1. SCD occurred in 4 patients (24%) (#3, #5, #6, and #16; Table 3), 2 of whom died despite an ICD. Notably, of those who experienced ventricular arrhythmia during or after GA, sevoflurane was used in at least 3, and 1 patient in particular has been arrhythmia-free in multiple other periods of GA by avoiding sevoflurane.

Four patients experienced SCD in this study. Patient #3 died at 1 month of life while still in the hospital after an episode of TdP. Patient #5, who had an ICD, developed refractory VF associated with deep hypoglycemia immediately after LCSD, which resulted in severe neurological damage and eventual discontinuation of treatment. Despite beta-blocker therapy and LCSD, this patient died of VF. Another patient (#6) initially had an ACA secondary to

TABLE 2 Genotypic Characteristics

Patient #	*CACNA1C* Mutation	Inheritance	Medical Therapy	Beta-Blocker Dose at Last Follow-Up	ICD	Outcome
1	Unknown	N/A	Nadolol	60 mg b.i.d.	Yes	Alive
2	p.Gly402Ser	De novo	Nadolol	40 mg once a day	Yes	Alive
3	p.Gly406Arg	De novo	Propranolol	5 mg/kg/day	No	Deceased
4	p.Gly406Arg	De novo	Atenolol	2 mg/kg/day	Yes	Alive
5	p.Gly406Arg	De novo	Propranolol, mexiletine	4 mg/kg/day	Yes	Deceased
6	p.Gly406Arg	De novo	Propranolol	4 mg/kg/day	No	Deceased
7	p.Gly406Arg	Paternal mosaicism	Nadolol	1 mg/kg/day	Yes	Alive
8	p.Ser405Arg	Unknown	Atenolol	2 mg/kg/day	No	Alive
9	p.Gly406Arg	Unknown	Propranolol	2 mg/kg/day	Yes	Alive
10	p.Gly406Arg	De novo	Propranolol, mexiletine	2 mg/kg/day	Yes	Alive
11	p.Ser405Arg	Unknown	Atenolol	2 mg/kg/day	Yes	Alive
12*	p.Gly402Ser	Unknown	Atenolol	2 mg/kg/day	Yes	Alive
13	p.Gly402Arg	De novo	Nadolol, mexiletine	N/A	Yes	Alive
14	p.Gly406Arg	Maternal mosaicism	Propranolol	3 mg/kg/day	Yes	Alive
15	p.Lys1211Glu	De novo	Propranolol	3 mg/kg/day	No	Alive
16	p.Gly406Arg	De novo	Propranolol	N/A	Yes	Deceased
17	p.Cys1021Arg	De novo	Propranolol	4 mg/kg/day	Yes	Alive

*Patient also had an additional KCNQ1 mutation (Ala590Thr) inherited from asymptomatic father.
b.i.d. = twice a day; ICD = implantable cardioverter-defibrillator; N/A = not available.

pulseless electrical activity, which was preceded by TWA during a pacemaker generator change under GA. The indication for permanent pacing was prolonged sinus pauses without 2:1 AV block. He was resuscitated successfully after 25 min with resultant significant anoxic neurological injury but did not have ventricular arrhythmias during the initial ACA event. The family did not agree to an ICD for this patient, who was then discharged home with an AED. One year later, after having made a nearly complete neurological recovery, the patient developed refractory VF while walking to the bathroom in the middle of the night and could not be successfully defibrillated with the family's AED. The fourth patient (#16) died after a hypoglycemic event despite an ICD. At the time of hypoglycemia, the patient had a seizure, but the ICD recording did not show a ventricular arrhythmia. However, in the emergency department, the patient developed intractable VF and could not be resuscitated.

DISCUSSION

Although the first patients with marked QT prolongation and syndactyly were described in the 1990s (14,15), it was only in 2004 when the molecular genetic mechanisms and terminology of the disorder were first established (1). The complex clinical constellation of TS can be attributed to the ubiquitous presence of $Ca_v1.2$ throughout the body and alterations in channel function. In addition, decreased calcium-dependent and voltage-dependent inactivation of the $Ca_v1.2$ channel caused by the TS-associated mutations, calcium-independent effects, such as transcriptional modification, may contribute to the noncardiac, phenotypic manifestations of the syndrome (16–18).

There are 3 main findings in this study. First, beta-blockers as standalone therapy may not be protective against SCD. Also, all beta-blockers may not have the same efficacy. Regarding the choice of beta-blockers, data from single-center studies and expert consensus state that nadolol is the preferred antiarrhythmic drugs for treatment of long QT syndrome (LQTS). In this study, of the 8 patients who experienced a cardiac arrest while taking beta-blockers, 1 was treated with atenolol, 3 with nadolol, and 4 with propranolol. Unfortunately, nadolol is not available worldwide, and this may limit its universal use as encountered in this international study. There are no randomized clinical trials addressing the comparative efficacy of different beta-blockers for treatment of LQTS, but experts concur that when nadolol is not been available or tolerated, treatment with propranolol is preferred to $beta_1$-selective agents such as atenolol (19).

Most patients were treated with a combination of antiarrhythmic agents and device therapy. At an average follow-up of 5 years, 77% of patients in our study were alive. These improved outcomes, compared with the initial report by Splawski et al. (1), may be attributed to early diagnosis and more

JACC: CLINICAL ELECTROPHYSIOLOGY VOL. 4, NO. 4, 2018
APRIL 2018:459–66

TABLE 3 Events Associated With ACA or SCD

Patient #	Mutation	Outcome	Rhythm at ACA/SCD	Associations
1	Unknown	Alive	TdP, VF	GA, nonexertional
2	p.Gly402Ser	Alive	TdP, VF	GA
3	p.Gly406Arg	Deceased	TdP, VF	Unknown
5	p.Gly406Arg	Deceased	TdP, VF	GA, nonexertional
6	p.Gly406Arg	Deceased	PEA, VF	GA, unknown
7	p.Gly406Arg	Alive	TdP	Nonexertional
9	p.Gly406Arg	Alive	TdP	GA
10	p.Gly406Arg	Alive	TdP	Fever, hypoglycemia
12	p.Ser405Arg	Alive	Likely TdP	GA
13	p.Gly402Ser	Alive	VF	Exertional, unknown
16	p.Gly406Arg	Deceased	Unknown	Hypoglycemia

ACA = aborted cardiac arrest; GA = general anesthesia; PEA = pulseless electrical activity; SCD = sudden cardiac death; TdP = torsades de pointes; VF = ventricular fibrillation.

aggressive medical and device therapy. Beta-blockers remain the mainstay of treatment, but mexiletine is an additional option for patients who continue to experience cardiac events while on beta-blockade. The evolution of smaller ICDs and alternative surgical implantation techniques has made it feasible to implant primary and secondary prevention ICDs in infants and young children, which was not possible in previous decades (20). Three patients also underwent LCSD, a procedure that has regained popularity in the past decade for primary and secondary prevention of malignant ventricular arrhythmias in LQTS (21,22). This remains an option for patients with TS, especially those with refractory symptoms.

The second finding of this study was that 8 of 11 ACA/SCD events (72%) were associated with either hypoglycemia or general anesthetic agents. It is known that patients with LQTS on beta-blocker therapy can develop episodes of hypoglycemia. We speculate that TS patients may be more prone to develop hypoglycemia due to potential episodic dysfunction of $Ca_v1.2$ signaling in the pancreatic beta-cells (1,4). This may be further complicated by treatment with beta-blockers, sometimes administered in high doses, which can mask early symptoms of hypoglycemia. Therefore, it would be prudent to avoid prolonged fasting in TS patients, and other strategies to prevent hypoglycemia should be investigated. GA, specifically with sevoflurane, was noted to be associated with ACA/SCD in several patients. Sevoflurane is known to prolong the QT interval even in normal children and may have further deleterious effects on patients with severe QT prolongation as seen in TS (23–25). Other contributory factors related to GA may include changes in sympathetic tone due to hypoxia, hypercarbia, local or light anesthesia, tracheal intubation, or emergence phenomena. Additionally, electrolyte imbalances and hypoglycemia may set in during the pre-procedural fasting period.

The third finding of this study was the prevalence of expanded TS-associated mutations within the *CACNA1C* gene and variable phenotypic features. Our study shows that the previously reported missense mutation p.Gly406Arg described in classic TS was present in only 10 patients (59%) (1–3). Three novel mutations were discovered in this case series (p.Ser405Arg, p.Gly402Arg, and p.Cys1021Arg). The clinical importance of this finding is that expanded genetic testing to the entire *CACNA1C*-encoded L-type calcium channel may be needed in patients with a TS-like phenotype if initial targeted testing of the hotspot exon8/8A is negative. Whether the variable TS phenotype is due to the type of mutation, its unique localization, alteration in signaling cascades, or other genetic/epigenetic factors is unresolved. Additionally, parental germline mosaicism was noted in 2 of our patients, which suggests some benefit for careful genotyping of the parent's DNA even for parents without signs of TS (11,26,27).

Finally, it should be noted that our case series consists of a heterogeneous cohort of TS patients. None of the patients in our case series were presented in the original reports by Splawski et al. (1,2). In contrast, the original report by Splawski et al (1) published in 2004 described a cohort of 17 TS patients with molecular homogeneity, and all patients had syndactyly in addition to multiple organ system involvement. This is often referred to as TS type 1 or classic TS in the literature. In 2005, Splawski et al. (2) described 2 additional cases with extreme QT prolongation and arrhythmias but with the absence of syndactyly. Both cases had de novo missense mutations affecting exon 8 (p.Gly406Arg or p.Gly402Ser) but not exon 8A, leading to the coinage of the term TS type 2. In the ensuing 12 years, additional mutations in the *CACNA1C* gene on exons outside the hotspot region of exon 8A/8 associated with variable phenotypic TS features have been reported, making the task of further TS classification more complex. It may be prudent to formulate the TS nomenclature on the basis of the susceptibility genes rather than individual mutations or phenotypic characteristics. Therefore, we propose that all TS phenotypes resulting from mutations in *CACNA1C* be classified as TS1. Because every TS patient may not be positive for the *CACNA1C* mutation, when the next TS susceptibility gene is discovered, it can be classified as TS type 2. It is also important to recognize that *CACNA1C* mutations may result in a cardiac phenotype of QT prolongation, hypertrophic cardiomyopathy, CHD, and premature SCD without involvement of other

organ systems. This should be annotated as cardiac-only TS (8). Another clarification needed is the terminology used for *CACNA1C* gene mutations resulting in isolated QT prolongation and arrhythmias without manifestation of other cardiac or extracardiac TS findings. Such cases should be simply referred to as LQTS type 8.

STUDY LIMITATIONS. Most notably, the small study cohort limits our ability to draw definitive conclusions regarding the different antiarrhythmic therapies prescribed. Given the retrospective nature of the study, clinical information on every aspect of multiorgan involvement in the cohort was not available. It is also possible that inclusion of novel *CACNA1C* mutations may have contributed to the positive outcomes in this cohort, although improvements in mortality were noted even among patients with the "classic" p.Gly406Arg mutations. Also, this study does not represent the complete list of TS patients diagnosed in the past decade. ICD complications were not encountered in this study, but this may be attributed to data collection bias and the small cohort of patients. In addition, although noncompliance with treatment with medications at the time of arrhythmic events was not reported by the treating physicians, the uniform compliance with treatment cannot be ascertained because of the intrinsic shortcomings of a retrospective study.

CONCLUSIONS

Our study shows that mortality in patients with TS is due to multifactorial mechanisms. Potential associations with sudden death events include hypoglycemia and specific general anesthetic agents. ICD therapy for primary and secondary prevention of SCD is frequently used for the management of TS with improved short-term survival. Due to the rarity of the disease, a worldwide prospective registry is needed for continued exploration into its clinical presentations, genetic mutations, malignant triggers, treatments, and outcomes.

ADDRESS FOR CORRESPONDENCE: Dr. Maully Shah, Perelman School of Medicine, University of Pennsylvania, The Children's Hospital of Philadelphia, 3401 Civic Center Boulevard, Philadelphia, Pennsylvania 19104. E-mail: shahm@email.chop.edu.

PERSPECTIVES

COMPETENCY IN MEDICAL KNOWLEDGE: TS is an extremely rare genetic disorder of the L-type cardiac channel $Ca_v1.2$ encoded by *CACNA1C*. The syndrome is characterized by multisystem abnormalities consisting of QT prolongation, CHD, syndactyly, facial dysmorphism, and developmental delay. Early presentation with heart block due to QT prolongation is frequently seen, and a high attrition rate due to SCD has been reported in previous case series.

TRANSLATIONAL OUTLOOK: A worldwide prospective registry is needed for genetic and clinical exploration of this rare syndrome with multisystem abnormalities. Future studies are needed to identify potential triggers for SCD and optimal treatment strategies to prevent SCD.

REFERENCES

1. Splawski I, Timothy KW, Sharpe LM, et al. Cav1.2 calcium channel dysfunction causes a multisystem disorder including arrhythmia and autism. Cell 2004;119:19–31.

2. Splawski I, Timothy KW, Decher N, et al. Severe arrhythmia disorder caused by cardiac L-type calcium channel mutations. Proc Natl Acad Sci U S A 2005;102:8089–96.

3. Hiippala A, Tallila J, Myllykangas S, et al. Expanding the phenotype of Timothy syndrome type 2: an adolescent with ventricular fibrillation but normal development. Am J Med Genet A 2015;167A:629–34.

4. Diep V, Seaver LH. Long QT syndrome with craniofacial, digital, and neurologic features: is it useful to distinguish between Timothy syndrome types 1 and 2? Am J Med Genet A 2015;167A:2780–5.

5. Boczek NJ, Miller EM, Ye D, et al. Novel Timothy syndrome mutation leading to increase in CACNA1C window current. Heart Rhythm 2015;12:211–9.

6. Wemhoner K, Friedrich C, Stallmeyer B, et al. Gain-of-function mutations in the calcium channel CACNA1C (Cav1.2) cause non-syndromic long-QT but not Timothy syndrome. J Mol Cell Cardiol 2015;80:186–95.

7. Betzenhauser MJ, Pitt GS, Antzelevitch C. Calcium channel mutations in cardiac arrhythmia syndromes. Curr Mol Pharmacol 2015;8:133–42.

8. Boczek NJ, Ye D, Jun F, et al. Identification and functional characterization of a novel CACNA1C-mediated cardiac disorder characterized by prolonged QT intervals with hypertrophic cardiomyopathy, congenital heart defects, and sudden cardiac death. Circ Arrhythm Electrophysiol 2015;8:1122–32.

9. Boczek NJ, Best JM, Tester DJ, et al. Exome sequencing and systems biology converge to identify novel mutations in the L-type calcium channel, CACNA1C, linked to autosomal dominant long QT syndrome. Circ Cardiovasc Genet 2013;6:279–89.

10. Fukuyama M, Wang Q, Kato K, et al. Long QT syndrome type 8: novel CACNA1C mutations causing QT prolongation and variant phenotypes. Europace 2014;16:1828–37.

11. Fröhler S, Kieslich M, Langnick C, et al. Exome sequencing helped the fine diagnosis of two siblings afflicted with atypical Timothy syndrome (TS2). BMC Med Genet 2014;15:48.

12. Liao P, Soong TW. CaV1.2 channelopathies: from arrhythmias to autism, bipolar disorder, and immunodeficiency. Pflugers Arch 2010;460:353–9.

13. Thornton PS, Stanley CA, De Leon DD, et al., Pediatric Endocrine Society. Recommendations from the Pediatric Endocrine Society for evaluation and management of persistent hypoglycemia

JACC: CLINICAL ELECTROPHYSIOLOGY VOL. 4, NO. 4, 2018
APRIL 2018:459–66

in neonates, infants, and children. J Pediatr 2015;167:238–45.

14. Marks ML, Whisler SL, Clericuzio C, et al. A new form of long QT syndrome associated with syndactyly. J Am Coll Cardiol 1995;25:59–64.

15. Reichenbach H, Meister EM, Theile H. The heart-hand syndrome. A new variant of disorders of heart conduction and syndactyly including osseous changes in hands and feet. Kinderarztl Prax 1992;60:54–6.

16. Phillip LR, Rodrriguez FH III Cardiac arrest refractory to standard intervention in atypical Timothy syndrome (LQT8 type 2). Proc (Bayl Univ Med Cent) 2016;29:160–2.

17. Krey JF, Pasca SP, Shcheglovitov A, et al. Timothy syndrome is associated with activity-dependent dendritic retraction in rodent and human neurons. Nat Neurosci 2013;16:201–9.

18. Hofmann F, Flockerzi V, Kahl S, et al. L-type CaV1.2 calcium channels: from in vitro findings to in vivo function. Physiol Rev 2014;94:303–26.

19. Ackerman MJ, Priori SG, Dubin AM, et al. Beta blocker therapy for long QT syndrome and catecholaminergic polymorphic ventricular tachycardia: are all beta-blockers equivalent? Heart Rhythm 2017;14:e41–4.

20. Cannon BC, Friedman RA, Fenrich AL, et al. Innovative techniques for placement of implantable cardioverter defibrillator leads in patients with limited venous access to the heart. Pacing Clin Electrophysiol 2006;29:181–7.

21. Schwartz PJ, Priori SG, Cerrone M, et al. Left cardiac sympathetic denervation in the management of high-risk patients affected by the long-QT syndrome. Circulation 2004;109:1826–33.

22. Bos JM, Bos KM, Johnson JN, et al. Left cardiac sympathetic denervation in long QT syndrome: analysis of therapeutic nonresponders. Circ Arrhythm Electrophysiol 2013;6:705–11.

23. Schneider AE, Bos JM, Ackerman MJ. Effect of left cardiac sympathetic denervation on the electromechanical window in patients with either type 1 or type 2 long QT syndrome: a pilot study. Congenit Heart Dis 2016;11:437–43.

24. Whyte SD, Booker PD, Buckley DG. The effects of propofol and sevoflurane on the QT interval and transmural dispersion of repolarization in children. Anesth Analg 2005;100:71–7.

25. Nathan AT, Antzelevitch C, Montenegro LM, Vetter VL. Case scenario: anesthesia-related cardiac arrest in a child with Timothy syndrome. Anesthesiology 2012;117:1117–26.

26. Etheridge SP, Bowles NE, Arrington CB, et al. Somatic mosaicism contributes to phenotypic variation in Timothy syndrome. Am J Med Genet A 2011;155A:2578–83.

27. Dufendach KA, Giudicessi JR, Boczek NJ, et al. Maternal mosaicism confounds the neonatal diagnosis of type 1 Timothy syndrome. Pediatrics 2013;131:e1991–5.

KEY WORDS **hypoglycemia, sudden cardiac death, syndactyly, Timothy syndrome, T-wave alternans, ventricular fibrillation**