Impact of Genetics on the Clinical Management of Channelopathies

Peter J. Schwartz, MD, Michael J. Ackerman, MD, PhD, Alfred L. George, Jr, MD, Arthur A. M. Wilde, MD, PhD

There are few areas in cardiology in which the impact of genetics and genetic testing on clinical management has been as great as in cardiac channelopathies, arrhythmic disorders of genetic origin related to the ionic control of the cardiac action potential. Among the growing number of diseases identified as channelopathies, 3 are sufficiently prevalent to represent significant clinical and societal problems and to warrant adequate understanding by practicing cardiologists: long QT syndrome, catecholaminergic polymorphic ventricular tachycardia, and Brugada syndrome. This review will focus selectively on the impact of genetic discoveries on clinical management of these 3 diseases. For each disorder, we will discuss to what extent genetic knowledge and clinical genetic test results modify the way cardiologists should approach and manage affected patients. We will also address the optimal use of genetic testing, including its potential limitations and the potential medico-legal implications when such testing is not performed. We will highlight how important it is to understand the ways that genotype can affect clinical manifestations, risk stratification, and responses to the therapy. We will also illustrate the close bridge between molecular biology and clinical medicine, and will emphasize that consideration of the genetic basis for these heritable arrhythmia syndromes and the proper use and interpretation of clinical genetic testing should remain the standard of care.

The discovery of the first 3 long QT syndrome (LQTS) susceptibility genes in 1995 and 1996 (1–3) had a transformative effect on the diagnosis and treatment of arrhythmias. It opened the way to the realization that molecular biology could no longer be regarded as “something weird, with an unfriendly jargon and of no interest for a clinician”; it allowed the understanding of how even simple amino acid substitutions (missense mutations) due to a single nucleotide substitution could produce significant functional alterations in cellular electrophysiology. In addition, by showing that most of the disease genes for a number of arrhythmic disorders of genetic origin were involved in the ionic control of the cardiac action potential, the discovery led to a description of these disorders as “cardiac channelopathies.” Other than some rare disorders, there are 3 truly important genetic heart rhythm diseases whose ignorance by practicing cardiologists could cost the lives of the patients seeking their medical advice. They are LQTS, catecholaminergic polymorphic ventricular tachycardia (CPVT), and Brugada syndrome (BrS).

This review will touch briefly on the main features of these potentially life-threatening yet highly treatable diseases (at least LQTS and CPVT), because many thorough clinical reviews are available (4–8). Instead, our focus will be centered selectively on discussing the impact exerted on clinical management by the progressive unraveling of the genetic mechanisms underlying these diseases. Specifically, we will examine for each of them whether, how, and to what extent genetic knowledge modifies clinical manifestations, risk stratification, and responses to therapy.
**Long QT Syndrome**

LQTS represents a leading cause of autopsy-negative sudden death in the young (9). It is characterized typically by a prolongation of the QT interval on electrocardiography (ECG) and by the occurrence of syncope or cardiac arrest, mainly precipitated by emotional or physical stress; however, some deaths occur when patients are at rest or asleep. LQTS includes the relatively common Romano-Ward (RW) variant, which has a prevalence of 1:2,000 live births (10), and the rare and extremely severe Jervell and Lange-Nielsen (JLN) syndrome accompanied by congenital deafness (11). The inheritance mode for RW is autosomal dominant or sporadic, whereas JLN shows autosomal recessive inheritance or sporadic cases of compound heterozygosity (11). LQTS contributes to sudden infant death syndrome (12) and even to stillbirths (13).

The ventricular tachyarrhythmia that underlies the cardiac events of LQTS is torsades de pointes (Tdp). This highly specific type of ventricular tachycardia is often self-limiting, thus producing transient syncope, but Tdp can also degenerate into ventricular fibrillation and cause cardiac arrest or sudden death. We still do not know why in certain patients Tdp stops after a few seconds, whereas in others it continues with devastating consequences.

The morphology of the T-wave is often useful for the diagnosis, and the lateral precordial leads are especially informative when they reveal biphasic or notched T waves (14). T-wave alternants in polarity or amplitude is a marker of major electrical instability and, when observed, is diagnostic (15). In the presence of syncopal episodes occurring under stressful conditions in an individual with marked prolongation of the QT interval, the diagnosis of LQTS is rather simple. For the more questionable situations, the so-called Schwartz score, originally proposed in 1993 (16) and updated more recently (6) (Table 1), can help. The standard and effective therapy for LQTS is based on β-blockers (propranolol and nadolol, whereas metoprolol has been linked to frequent recurrences [17]), which should be administered to still asymptomatic patients with QT prolongation, given the potential for sudden death as the first disease manifestation. For patients receiving full-dose β-blocker therapy in the case of a first recurrence of syncope, left cardiac sympathetic denervation (18,19) is the treatment of choice, whereas in the case of cardiac arrest, recurrent syncope, or signs of very high risk, an implantable cardioverter defibrillator (ICD) is appropriate (8,20).

### Updated Genetics

Sixteen genes have been identified so far as responsible for or associated with LQTS (Table 2). The 3 main genes, KCNQ1 (LQT1), KCNH2 (LQT2), and SCN5A (LQT3), account for approximately 75% of clinically definite LQTS, whereas the minor genes contribute an additional 5% collectively. An estimated 20% of LQTS remains genetically elusive.

*KCNQ1* encodes the α-subunit of the K+ channel Kv7.1, generating I\(_{Ks}\), which is physiologically increased by sympathetic activation and is essential for QT adaptation during heart rate increases. When I\(_{Ks}\) is diminished or dysfunctional, the QT interval fails to shorten appropriately during tachycardia, thus leading to a potentially arrhythmogenic condition. Heterozygous *KCNQ1* mutations cause the dominant RW LQTI syndrome and is the most common LQTS genotype, accounting for 30% to 35% of LQTS.
Homozygous mutations in \( KCNQ1 \), or compound heterozygous mutations, can cause the autosomal recessive JLN variant. Different effects may be produced by mutations in this multimeric \( K^+ \) channel. If the mutation prevents the co-assembly such that only the wild-type subunit can tetramerize, then a mechanism of haploinsufficiency whereby \( I_{\text{KS}} \) is reduced by 50% emerges. On the other hand, if the mutant-containing allele can tetramerize and "poison" the tetramer, then a dominant negative mechanism emerges, resulting in a minimum residual of 6% current density. The dominant-negative effect of certain \( KCNQ1 \) mutations may manifest as a failure to modulate \( I_{\text{KS}} \) by \( \beta \)-adrenergic signaling (21,22).

The second most common gene harboring LQTS mutations is \( KCNH2 \), encoding HERG, the \( \alpha \)-subunit of the \( I_{\text{Kr}} \) current.

### Table 2: Molecular Basis of Cardiac Channelopathies

<table>
<thead>
<tr>
<th>Gene</th>
<th>Locus</th>
<th>Protein</th>
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<tbody>
<tr>
<td><strong>LQTS</strong></td>
<td></td>
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<tr>
<td>Major LQTS genes</td>
<td></td>
<td></td>
</tr>
<tr>
<td>( KCNQ1 ) (LQT1)</td>
<td>11p15.5</td>
<td>( I_{\text{Kv}} ) potassium channel alpha subunit (KVLQT1, ( K_\text{V}7.1 ))</td>
</tr>
<tr>
<td>( KCNH2 ) (LQT2)</td>
<td>7q35-36</td>
<td>( I_{\text{Kr}} ) potassium channel alpha subunit (HERG, ( K_\text{V}11.1 ))</td>
</tr>
<tr>
<td>SCN5A (LQT3)</td>
<td>3p21-p24</td>
<td>Cardiac sodium channel alpha subunit (( Na_\text{s,1.5} ))</td>
</tr>
<tr>
<td><strong>Minor LQTS genes</strong></td>
<td></td>
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<tr>
<td>(listed alphabetically)</td>
<td></td>
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</tr>
<tr>
<td>AKAP9</td>
<td>7q21-q22</td>
<td>Yotiao</td>
</tr>
<tr>
<td>CACNA1C</td>
<td>12p13.3</td>
<td>Voltage gated L-type calcium channel (( Ca_{\text{v},1.2} ))</td>
</tr>
<tr>
<td>CALM1</td>
<td>14q32.11</td>
<td>Calmodulin 1</td>
</tr>
<tr>
<td>CALM2</td>
<td>2p21.3-p21.1</td>
<td>Calmodulin 2</td>
</tr>
<tr>
<td>CAV3</td>
<td>3p25</td>
<td>Caveolin-3</td>
</tr>
<tr>
<td>KCNE1</td>
<td>21q22.1</td>
<td>Potassium channel beta subunit (MinK)</td>
</tr>
<tr>
<td>KCNE2</td>
<td>21q22.1</td>
<td>Potassium channel beta subunit (MIRP1)</td>
</tr>
<tr>
<td>KCN5</td>
<td>11q24.3</td>
<td>Kir3.4 subunit of ( I_{\text{KACs}} ) channel</td>
</tr>
<tr>
<td>SCN4B</td>
<td>11q23.3</td>
<td>Sodium channel beta 4 subunit</td>
</tr>
<tr>
<td>SNTA1</td>
<td>20q11.2</td>
<td>Syntrophin-alpha 1</td>
</tr>
<tr>
<td><strong>Andersen-Tawil Syndrome</strong></td>
<td></td>
<td></td>
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<tr>
<td>( KCN2 ) (ATS1)</td>
<td>17q23</td>
<td>( I_{\text{K1}} ) potassium channel (( K_\text{r,2.1} ))</td>
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<tr>
<td><strong>Ankyrin-B Syndrome</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ANKB</td>
<td>4q25-q27</td>
<td>Ankyrin B</td>
</tr>
<tr>
<td><strong>Timothy Syndrome</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CACNA1C (TS)</td>
<td>12p13.3</td>
<td>Voltage gated L-type calcium channel (( Ca_{\text{v},1.2} ))</td>
</tr>
<tr>
<td><strong>Catecholaminergic polymorphic ventricular tachycardia</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RYR2 (CPVT1)</td>
<td>1q42.1-q43</td>
<td>Ryanodine receptor 2</td>
</tr>
<tr>
<td>CASQ2 (CPVT2)</td>
<td>1p13.3</td>
<td>Calsequestrin 2</td>
</tr>
<tr>
<td>( KCN2 ) (CPVT3)</td>
<td>17q23</td>
<td>( I_{\text{K1}} ) potassium channel (( K_\text{r,2.1} ))</td>
</tr>
<tr>
<td>CALM1</td>
<td>14q32.11</td>
<td>Calmodulin 1</td>
</tr>
<tr>
<td>TRDN</td>
<td>6q22.31</td>
<td>Triadin</td>
</tr>
<tr>
<td><strong>Brugada Syndrome</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SCN5A (BrS1)</td>
<td>3p21-p24</td>
<td>Cardiac sodium channel alpha subunit (( Na_\text{s,1.5} ))</td>
</tr>
<tr>
<td><strong>Minor BrS genes</strong></td>
<td></td>
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<td>(listed alphabetically)</td>
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</tr>
<tr>
<td>CACNA1C</td>
<td>2p13.3</td>
<td>Voltage gated L-type calcium channel (( Ca_{\text{v},1.2} ))</td>
</tr>
<tr>
<td>CACNA2D1</td>
<td>7q21-q22</td>
<td>Voltage gated L-type calcium channel 2 delta 1 subunit</td>
</tr>
<tr>
<td>CACNB2</td>
<td>10p12</td>
<td>Voltage gated L-type calcium channel beta 2 subunit</td>
</tr>
<tr>
<td>DLG1</td>
<td>3q29</td>
<td>Synapse-associated protein 97</td>
</tr>
<tr>
<td>GDPD1L</td>
<td>3p22.3</td>
<td>Glycerol-3-phosphate dehydrogenase 1-like</td>
</tr>
<tr>
<td>HCN4</td>
<td>15q24.1</td>
<td>Hyperpolarization-activated cyclic nucleotide-gated channel 4</td>
</tr>
<tr>
<td>KCND3</td>
<td>1p13.2</td>
<td>Voltage-gated potassium channel (( I_{\text{K1}} ) subunit K\text{v,4.3}</td>
</tr>
<tr>
<td>KCNE3</td>
<td>11q13.4</td>
<td>Potassium channel beta subunit 3 (MIRP2)</td>
</tr>
<tr>
<td>KCNE5</td>
<td>Xq22.3</td>
<td>Potassium channel beta subunit 5</td>
</tr>
<tr>
<td>KCN8</td>
<td>12p12.1</td>
<td>Inward rectifier ( K^{\text{+}} ) channel Kir6.1</td>
</tr>
<tr>
<td>M0G1</td>
<td>17p13.1</td>
<td>RAN guanine nucleotide release factor 1</td>
</tr>
<tr>
<td>SCN1B</td>
<td>19q13</td>
<td>Sodium channel beta 1</td>
</tr>
<tr>
<td>SCN3B</td>
<td>11q24.1</td>
<td>Sodium channel beta 3</td>
</tr>
<tr>
<td>SLMAP</td>
<td>3p14.3</td>
<td>Sarcolemma-associated protein</td>
</tr>
</tbody>
</table>

BrS = Brugada syndrome; LQTS = long QT syndrome.
components of the delayed rectifier \( I_{K} \) current, the major determinant of phase 3 repolarization in ventricular cardiomyocytes. LQT2-causative mutations in \( \text{SCN}2A \) provoke a reduction in \( I_{K} \) current, most commonly by mechanisms involving impaired trafficking of the protein to the plasma membrane (23).

The third major LQTS gene is \( \text{SCN}5A \), which encodes the \( \alpha \)-subunit of the cardiac sodium channel (\( \text{Na}_{v}1.5 \)) that conducts the depolarizing inward sodium current. A few months after its identification as an LQTS gene in 1995 (1), it was shown that the \( \text{SCN}5A-\Delta \text{KPQ} \) mutation produces the LQTS phenotype by increasing the persistent (or late) \( \text{Na}^{+} \) inward current and, therefore, prolonging action potential duration (24). This study provided the first evidence linking a mutation to a functional alteration in the ionic control of ventricular repolarization and paved the way to all subsequent functional studies that have become the gold standard to establish that a novel mutation in a patient with LQTS is likely a disease-causing one.

After the identification of the first 3 major LQTS genes (1–3), several others were identified. \( \text{KCNE}1 \) and \( \text{KCNE}2 \) encode \( K^{+} \) channel auxiliary subunits that are associated with the \( \alpha \)-subunits encoded by \( \text{KCNQ}1 \) and \( \text{KCNH}2 \). Mutations in \( \text{KCNE}1 \) may cause the dominant RW (LQTS) or, if present in homozygosity or compound heterozygosity, the recessive JLN syndrome (11). There are few cases of \( \text{KCNE}2 \) mutations associated with LQTS, and most of them represent acquired LQTS associated with specific drugs, almost all \( I_{K} \) blockers.

Among the sodium channel interacting protein-coding genes, \( \text{C} \alpha \text{V}3 \) (25), \( \text{SCN}4B \) (26), and \( \text{SNTA}1 \) (27) are regarded as additional LQTS genes (LQT9, LQT10, and LQT12) that essentially mimic LQT3. The AKA9-encoded Yotiao is involved in the phosphorylation of Kv7.1, and its mutation has been described in LQT11, which functionally mimics LQT1. Two missense mutations in \( \text{CACNA}1C \), encoding a voltage-gated calcium channel, are linked to Timothy syndrome (LQT8), a rare and extremely malignant LQTS variant. In a large Chinese family, a heterozygous mutation was identified in the inward rectifying \( K^{+} \) channel subunit Kir3.4, encoded by \( \text{KCNJ}5 \). The variant was present in all the 9 affected family members and was absent in >500 ethnically matched controls, suggesting a role in the pathogenesis of LQT13. On the other hand, the \( \text{ANKB} \), \( \text{KCNJ}2 \), and \( \text{CACNA}1C \) genes, often referred to as LQT4, LQT7, and LQT8, are associated with complex clinical disorders: ankyrin-\( B \) syndrome, Andersen-Tawil syndrome, and Timothy syndrome, respectively. In the first two, prolongation of the QT interval is modest. Until LQTS-causing mutations are found in these genes in patients with clinically definite LQTS, these 3 genes should not be strictly considered as part of LQTS.

A most malignant form of LQTS that causes recurrent cardiac arrest due to ventricular fibrillation manifesting in infancy has been found to be associated with mutations in \( \text{CALM}1 \) and \( \text{CALM}2 \), 2 of the 3 human genes encoding calmodulin (28). Calmodulin is a ubiquitous multifunctional \( \text{Ca}^{2+} \) binding protein, and overexpression of calmodulin mutants with defective \( \text{Ca}^{2+} \) binding produces major prolongation of ventricular action potentials (29,30). In 2 unrelated infants with QT prolongation and very early occurrence of cardiac arrest due to ventricular fibrillation, whole exome sequencing revealed de novo mutations in \( \text{CALM}1 \) or \( \text{CALM}2 \). A subsequent candidate gene screening in a cohort of 82 LQTS genotype-negative subjects identified 2 more \( \text{CALM}1 \) mutation carriers (28). All 4 patients share strikingly similar clinical manifestations: major QT prolongation (all >600 ms), T-wave alternans, cardiac arrest in infancy, multiple episodes of ICD-terminated ventricular fibrillation mostly triggered by sympathetic activation, and poor response to pharmacological and nonpharmacological interventions.

### Genetics and Arrhythmia Triggers

A study performed on approximately 700 patients of known genotype and all with arrhythmic events demonstrated that the triggers for arrhythmias in LQTS are gene-specific (31). Patients with LQT1 are at risk especially during sympathetic activation, as with physical exercise or emotional stress (Fig. 1), and this stems from the fact that they have a lower-than-normal \( I_{K} \), and therefore their ability to shorten the QT interval when heart rate increases is impaired. Patients with LQT2 and LQT3, who have a normal level of \( I_{K} \), are at no special risk during physical exercise and sport activity. Patients with LQT2 are exquisitely sensitive to sudden noises, such as alarm clocks or telephone ringing, whereas patients with LQT3 tend to have their events while at rest or while asleep. In the large study by Schwartz et al. (31), 99% of

![Figure 1](http://content.onlinejacc.org/)  
**Triggers for Fatal Cardiac Events in Patients With LQT1, LQT2, and LQT3**  
**Arrows** point out the rare occurrence of these events during sympathetic activation in patients without mutations affecting the \( I_{K} \) current. Modified with permission from Schwartz et al. (31). LQTS = long QT syndrome; SCD = sudden cardiac death.
the events that occurred while swimming were in patients with LQT1 and 80% of the events triggered by sudden noises were in patients with LQT2, thus allowing the shrewd clinician to suspect the correct genotype on the basis of simple clinical history and well before obtaining the genetic results.

Genetics and Risk Stratification

Since the early days of molecular genetics for LQTS, attempts have been made to correlate genotypes with outcomes. The first large study reported on 647 patients with LQTS and suggested interactions among genotype, QTc, and gender (32). The risk of cardiac events, higher for female patients with LQT2 and male subjects with LQT3, increased in the presence of marked QT prolongation (QTc >500 ms). Patients with LQT1 were less likely to experience events, probably because of the high percentage (36%) of patients with the disease-causing mutation but with a QTc <440 ms. The existence of these genotype-positive/phenotype-negative patients is related to the low penetrance existing in LQTS, which was postulated in 1980 (33) and demonstrated in 1999 (34).

A significant step forward came with the realization that in addition to genotype-based risk stratification, intragenic risk stratification was possible for LQT1 and LQT2 based on molecular/structural location and cellular function. In 2002 (35) and 2007 (36), Moss et al indicated first that patients with LQT2 with pore-localizing mutations are at higher risk and second that in patients with LQT1 both the transmembrane location of the mutations and their dominant-negative effect are independent risk factors for cardiac events. These studies indicated that not all mutations on the same gene produce a similar clinical phenotype and initiated a series of intriguing revelations on the complexity of the genotype–phenotype correlation. An evolution of these studies led to the realization that there are areas in the genes, such as the Kv7.1 cytoplasmic loops, associated not only with higher arrhythmic risk but also with a particularly good response to β-blocker therapy (21).

However, neither the localization of a mutation nor its cellular electrophysiological effect is sufficient to consistently predict the impact on clinical manifestations. The most striking example of mutation-specific behavior is probably that of KCNQ1-A341V, a relative hotspot mutation characterized by unusual clinical severity demonstrated by 80% of the patients being symptomatic, with >30% experiencing cardiac arrest or sudden death (37,38). What is puzzling is that A341V is only a mildly dominant negative mutation producing a relatively modest IKs loss.

Genetics, Response to Therapy, and Clinical Management

Since the identification of the first LQTS genes, high hopes were generated that understanding the molecular underpinnings of the disease would inspire novel therapeutic approaches. So far, this has been only partially true. Still, progress in the management of these patients based on genotype–phenotype studies is impressive and undeniable, and so far, the diagnostic, prognostic, and therapeutic effects of genetic testing have been realized most fully for LQTS compared with all other genetically mediated channelopathies and cardiomyopathies (7).

The response to β-blocker therapy is in part gene-specific, but not as much as previously thought. Clearly, β-blockers are extremely effective for patients with LQT1 (31,39,40) and also effective for patients with LQT2, but female patients with LQT2 are less fully protected. Contrary to some previous opinion, largely dependent on the inclusion in the analysis of a subgroup largely unresponsive to therapy and represented by patients with events in the first year of life (41), β-blockers are effective also for patients with LQT3 as indicated by a study in >400 patients (42).

Within months after the cellular demonstration, in August 1995, that the electrophysiological consequence of an SCN5A mutation is an increase in persistent Na+ current (24), clinical (43) and experimental (44) evidence was provided that a sodium channel blocker, mexiletine, could markedly shorten the QT interval in patients with LQT3, but not in patients with LQT1 and LQT2, and block the persistent Na+ current. It was immediately warned that mexiletine should have never been used instead of β-blockers but as a potentially useful addition. Indeed, despite mexiletine-mediated clear evidence of benefit in certain patients, there have been failures in others. The response to Na+ channel blockers is clearly mutation-specific, and this dictates the correct clinical approach: to always test the QT-shortening effect of mexiletine using the acute oral drug testing approach (45) with one-half of the daily dose while monitoring the patient’s ECG for 2 h; if the QTc shortens by more than 40 ms without undue PR lengthening, then it is reasonable to add mexiletine to the β-blocker therapy. There is current interest as potential LQT3-specific therapy in ranolazine, a drug more selective for the persistent Na+ current, but the available clinical data are scant and short term (46). It should not be forgotten that Na+ channel blockers have the potential to impair cardiac conduction, and vigilant ECG monitoring is necessary when patients with LQT3 are treated with mexiletine to avoid serious consequences. With specific mutations, mexiletine and propranolol may have beneficial synergistic effects in correcting major electrophysiological abnormalities (47). Flecainide should be considered for patients with LQTS because of its IKc blocking effect.

The major impact of genetics has been in the general management of the patients and their families. As to the patients, the identification of specific triggers for the arrhythmic events (31) has led to rational attempts to avoid or counter the at-risk situations. Those with LQT1 are advised to avoid excessive stress, be it physical or mental, and specific activities such as swimming unless under proper
protection. For those with LQTS2, it is important to minimize sudden noises, especially when resting; this means to avoid telephones and alarm clocks in the bedroom and to wake up the affected children without yelling. Also, because sleep deprivation and disruption are particularly bad for women with LQTS2 in the postpartum period, it is advisable that fathers find the proper way to feed their infants at nighttime without waking the mothers. Patients with LQTS2 and LQTS3, because of their “normal” Ik, are not expected to be at special risk during physical activity.

As to the families, the issue is that of “cascade screening” (48), that is, once the disease-causing mutation is identified in the proband, the entire family should undergo testing for that specific mutation, which is rapid and inexpensive, to identify the mutation carriers with normal QT. This concept is the direct consequence of the fact that low penetrance is common in LQTS (34), which confirms the original hypothesis (33) that some patients may be affected by LQTS and nonetheless have a normal QT interval. This implies that normal findings on an ECG cannot be used to exclude LQTS and mandates the necessity to perform molecular screening in all family members once the disease-causing mutation has been identified in the proband.

Cascade screening allows the identification, as mutation positive, of individuals who would have been otherwise considered unaffected and therefore would have remained at risk for potentially life-threatening arrhythmias as a spontaneous event or more likely as provoked by a variety of drugs with an Ik,-blocking effect. This leads to prophylactic treatment in >70% of mutation-positive individuals (49). Also important is the fact that those family members who are found to be negative for the family’s disease-causing mutation will be relieved to learn that they are not at risk and that they should not fear for their offspring.

The magnitude of the impact that genetic screening has on clinical cardiology is exemplified by the fact that not performing cascade screening could lead to a number of otherwise avoidable deaths among those genotype-positive/ phenotype-negative family members of affected patients. These are deaths that could be prevented partly by therapy, when appropriate, and partly by providing on a regular basis an updated list of drugs to carefully avoid. Cascade screening forcefully demonstrates that molecular biology and genetics can no longer be regarded as tools for researchers, but nowadays represent an essential component of good medical care.

### Catecholaminergic Polymorphic Ventricular Tachycardia

Akin to LQT1, CPTV is characterized phenotypically by exercise-induced syncop, seizures, or sudden death in the setting of a structurally normal heart (50–52). However, in contrast to LQTS, the ECG at rest is typically normal with only subtle, nondiagnostic bradycardia and U waves occasionally present. Instead, provocative stress testing by treadmill, cycle, or isoproterenol is the key diagnostic test to elicit CPVT’s trademark signature of exercise-induced bidirectional ventricular tachycardia. Of note, although fairly specific for CPVT, exercise-induced bidirectional ventricular tachycardia is insensitive because the majority of patients with mutation-proven CPVT do not manifest this arrhythmia (52). Moreover, patients with CPVT1 with a negative exercise stress test are not at zero risk (52). Rather, in the context of a positive personal or family history, CPVT should be suspected when a stress test exhibits the onset of premature ventricular contractions (PVCs) when the heart rate reaches approximately 110 to 130 beats/min. At this workload, this exercise-induced ectopy will commence with single, intermittent PVCs and progress typically to PVCs in bigeminy and couplets. Only occasionally will more complex ectopy ensue. Often the exercise-induced ectopy will burn out (return to normal sinus rhythm) at the highest work load/peak heart rate, and normal sinus rhythm almost always persists throughout the recovery phase.

Because CPVT is more arrhythmic than LQTS with higher estimated fatality and higher breakthrough rates during conventional β-blocker therapy, it is critical to distinguish CPVT from LQTS (53). Patients with CPVT often have been misdiagnosed as having “atypical” LQTS (54). As a diagnostic pearl, in the setting of an exercise-triggered cardiac event, a resting QTc <460 ms, and a structurally normal heart, CPVT rather than “concealed” or “normal QT interval” LQTS is far more likely to be the root cause.

Pathogenetically, approximately 50% to 60% of CPVT stems from heritable or sporadic mutations in the RYR2-encoded cardiac ryanodine receptor/calcium release channel, a critical regulator of intracellular calcium (55). RYR2 is one of the largest genes in the human genome, with its 105 translated exons, that encodes for a protein containing 4,967 amino acids. However, there are 3 particular domains/clusters encoded by 16 exons where two-thirds of the current CPVT1-associated mutations localize and all published mutations currently reside within less than half of RYR2’s exons (56,57).

In addition to CPVT1, rare autosomal recessive subtypes of CPVT stem from mutations in CASQ2-encoded calquestrin 2 (CPVT2) (58) or TRDN encoding the junctional protein triadin (59) (CPVT4). Mutations in CALM1 encoding calmodulin were discovered recently in 1 family with autosomal dominant CPVT-like phenotype and in a de novo single case, all genotype-negative for RYR2 or CASQ2 mutations (60) (CPVT5). Also, mutations in the KCNJ2-encoded Kir2.1 can express a clinical phenotype that mimics autosomal dominant CPVT (61) (CPVT3). Generally, loss-of-function KCNJ2 mutations cause type 1 Andersen-Tawil syndrome (ATS1), a heritable channelopathy readily distinguishable from CPVT with characteristic U waves, facial/skeletal stigmata, and a more benign prognosis. However, several KCNJ2 mutations have been identified in patients clinically diagnosed with CPVT because of complex
ventricular ectopy including bidirectional ventricular tachycardia with no clinical features to suggest ATS1.

CPVT genetic testing is recommended for any patient in whom a cardiologist has established a clinical index of suspicion for CPVT based on the basis of examination of the patient’s clinical history, family history, and expressed ECG phenotype during provocative stress testing with cycle, treadmill, or catecholamine infusion. Mutation-specific genetic testing is recommended for family members and appropriate relatives after the identification of the CPVT-causative mutation in an index case (62). Presently, the primary purpose of CPVT genetic testing is diagnostic, to genetically confirm a clinically suspected case of CPVT, establish the particular genotype, and identify the potentially at-risk relatives (63).

However, in stark contrast to the genotype-specific, region-specific, and even mutation-specific risk-stratifying information that has emerged in LQTS, no definitive domain-specific or mutation-specific prognostication can be made for RYR2-mediated CPVT (i.e., CPVT1). Preliminary data suggest that relatives carrying an RYR2 mutation in the C-terminal channel-forming domain may be at greater arrhythmic risk than those with a mutation in the N-terminal domain (64).

Thus, all patients with clinically manifest CPVT1 are treated on the basis of their phenotype without regard to any details about the particular mutation. Here, the genotype does not guide therapy. Phenotype-guided therapy generally consists of β-blocker therapy and/or left cardiac sympathetic denervation therapy, and if necessary, combination therapy with the addition of flecainide (53,65–68). Device therapy with an implantable defibrillator should be the last intervention (rather than the observed all too often first one) for only the highest-risk subjects with CPVT because of the uncommon but concerning issue of a CPVT-related ICD storm, whereby the ICD ultimately fails to rescue the patient (69,70).

Although the chief purpose of CPVT genetic testing is diagnostic rather than prognostic or therapeutic, the genotype influences the management and treatment of a patient with genetically confirmed CPVT in 2 important ways. First, it is important to distinguish KCNJ2-mediated CPVT (CPVT3) from the more common CPVT1 because the treatment strategy is different for the 2 genotypes. In contrast to the phenotype-guided treatment strategy for CPVT1 or genotype-negative/phenotype-positive CPVT, patients with KCNJ2-mediated CPVT may be more responsive to primary therapy with flecainide or mexiletine rather than β-blocker therapy (71). In addition, the protective, antiarrhythmic effect of LCSD has been demonstrated more clearly for patients with CPVT1 than for patients with CPVT3 (65).

Second, mutation-specific confirmatory testing in relatives enables prophylactic β-blocker therapy to be initiated at a young age if deemed necessary (64). Without the genotype, potentially at-risk family members would be revealed only after they are old enough to undergo a provocative stress test or if they manifest a concerning symptom. Considering that the first concerning symptom can be sudden death and that up to 15% of autopsy-negative unexplained sudden death subjects are CPVT1 positive (72), identifying a potentially vulnerable CPVT1-positive relative as early as possible is an indirect but potentially lifesaving therapeutic contribution of CPVT genetic testing.

Brugada Syndrome

BrS is a hereditary disease characterized by its “signature sign”, a coved-type ST-segment elevation in the anterior precordial leads (V₁ to V₃), referred to as a “type 1 Brugada ECG pattern”, and by the presence of right ventricular conduction abnormalities and life-threatening ventricular arrhythmias (73,74). The typical case is a 40-year-old resuscitated man without clear evidence for structural heart disease and with a family history for (nocturnal) sudden cardiac death. Indeed, up to 75% of those clinically affected are of male gender, and the mean age of onset of events is approximately 40 years, but with a wide range. A family history of sudden cardiac death is reported in 20% to 50% of cases. There is an autosomal dominant pattern of transmission, with highly variable and often low penetrance. Several aspects are still unclear, especially the pathophysiology of the right precordial ST-segment elevation (75).

BrS is a genetically heterogeneous disease, with the involvement of at least 13 different genes (76,77). Most mutations occur in genes with an impact on the function of cardiac Na⁺ channels. SCN5A, the gene encoding for the β-subunit of the cardiac sodium channel, is involved in 20% to 25% of patients. More than 200 SCN5A BrS-related mutations have been described to date (78). All mutations induce a reduction in the sodium current amplitude and do so through several mechanisms, including altered channel kinetics (e.g., faster inactivation or slower recovery from inactivation), trafficking defects, and generation of truncated proteins. A pure, self-sufficient causative role of loss-of-function SCN5A mutations has been challenged because in several large SCN5A-related BrS families, affected individuals did not carry the presumed familial disease-causing mutation, thus suggesting that SCN5A may actually represent a strong modifier (79).

Other genes with an impact on sodium channel function are the sodium channel β-subunit genes (SCN1B, SCN3B), affecting channel kinetics; glycerol-3-phosphate dehydrogenase 1-like enzyme (GPD1L); MOG1; and SLMAP, which affects trafficking of sodium channels. Potentially causative variants are also found in the calcium channel genes (CACNA1C, CACNB2B, CACNA2D1), in genes that affect the transient outward current (Iₒ) (KCNE3, KCND3, KCNE5), and in the gene that forms the pore-forming unit of the adenosine triphosphate-sensitive potassium current (IₖATP KCNJ8) (76). Involvement of most of these genes has been described in single patients and families with BrS,
Although mutations in \textit{CACNA1C} and \textit{CACNB2B} are reported to contribute to up to 11% of BrS. In basic electrophysiological studies, the calcium channel genes lead to loss of function of basal L-type calcium current (I\textsubscript{Ca,L}); a mutation in \textit{KCNE3}, \textit{KCND3}, or \textit{KCNE5} leads to a gain of function of I\textsubscript{Ks}; and mutations in \textit{KCNJ8} increase I\textsubscript{KATP}.

Genotype–phenotype correlation studies in BrS are sparse. Initial studies indicate that \textit{SCN5A}-associated BrS typically presents with longer conduction intervals in all cardiac compartments (80). Meta-analyses consistently show that the presence or absence of an \textit{SCN5A} mutation does not affect clinical outcome (81). However, within the \textit{SCN5A} cohort, the type of \textit{SCN5A} mutation may be useful for risk stratification, with nonsense mutations giving rise to truncated protein, leading to more severe conduction disorders and more symptoms (82). Calcium channel–related BrS seems to associate with shorter-than-normal QTc intervals, but it is not clear whether this affects prognosis (83).

Because the diagnosis of BrS is made on clinical grounds, genetic testing is not required for this goal. Yet, the finding of a loss-of-function \textit{SCN5A} mutation might help in a clinically uncertain diagnosis. As indicated earlier, knowledge of a mutation does not affect prognosis, with the possible exception of specific findings in \textit{SCN5A}. BrS genetic testing can be useful for any patient in whom there is reasonable suspicion for BrS based on the examination of the patient’s and his/her family’s clinical history, a clear ECG phenotype based on resting 12-lead ECGs, and/or provocative drug challenge testing. Because the presence of a disease-causing mutation does affect lifestyle (e.g., fever and specific drugs should be avoided) (84), cascade genetic screening is recommended for family members after the identification of the BrS-causative mutation in an index case. In the setting of an isolated type 2 or 3 Brugada ECG pattern, genetic testing has no place (61).

**Issues With Genetic Testing in Channelopathies**

Advances in deciphering the molecular basis for heritable cardiac arrhythmia susceptibility have reshaped the diagnostic paradigm and clinical management of these 3 familial arrhythmia syndromes (cardiac channelopathies) to include genetic testing, gene-specific considerations in therapy, and increased awareness of the need to assess disease risk in family members. The accurate ascertainment of family history, the proper use and interpretation of genetic test results, and the identification and management of at-risk family members have become the standard of care in this field.

**Optimal use of genetic testing.** Ascertainment of a comprehensive family history of cardiac arrhythmias and unexpected death is of utmost importance when considering the diagnosis of a heritable arrhythmia syndrome. In performing a detailed family history, special attention should be paid to the occurrence of syncope or unexpected sudden death, especially among young adults and children in the family, noting special circumstances surrounding unexpected death (e.g., drowning, seizures) and identifying any relatives with implanted cardiac devices along with the indication. A pedigree drawing is essential for the recognition of a mode of inheritance compatible with a monogenic disorder (e.g., autosomal dominant, autosomal recessive, X-linked). Information about the patient and close relatives is ultimately helpful in making final decisions about the use of genetic testing to confirm clinical suspicions. However, incomplete penetrance or subclinical disease expression may obscure the pattern of disease segregation in a family. This is unfortunately a common problem in families with BrS because of low penetrance (79) and may also obfuscate the recognition of inheritance patterns in LQTS (31). Severe and early-onset forms of LQTS and other syndromes especially may be caused by de novo mutations, in which case no family history is expected (47,85,86).

Genetic testing is a specialized diagnostic procedure available for LQTS, BrS, and CPVT through commercial and research laboratories (87). In the United States, clinical genetic testing laboratories must meet stringent criteria for quality standards that conform to the federal Clinical Laboratory Improvement Amendments passed in 1988 (88). Further, unlike more commonly used laboratory tests, genetic testing should be performed after the patient is informed about the potential risks, benefits, and limitations. Involvement of a genetic counselor is ideal in circumstances in which physician time or knowledge is limited. Despite these requirements, genetic testing can have tremendous value in identifying mutations that help confirm clinical suspicions, select genotype-specific therapy, and direct specific testing of at-risk relatives.

**Pitfalls and limitations of genetic testing.** The current yields of genetic testing for each of these syndromes range from 25% (BrS) to 80% (LQTS). Further, the methods to identify mutations are not 100% sensitive, and therefore a negative genetic test cannot exclude the disorder by itself. Also, certain detectable DNA sequence variants may not have a clear causal role in a patient’s condition because they are rare polymorphisms (89) or located in regions of the channel protein that have unknown functional importance. On the basis of several years of genetic testing experience in the academic and commercial sectors, we now know that most mutations are “private” (i.e., occurring in a single family) missense mutations with uncertain functional or pathophysiological consequences (77,90). Therefore, interpreting genetic test results is often confounded by the discovery of “variants of unknown significance” for which there are insufficient data or predictive tools to assess accurately the likelihood that a particular variant predisposes to an arrhythmia or whether the change is merely a benign rare variant (91). Only a small fraction of all identified genetic variants in the myriad genes associated with LQTS, BrS, and CPVT have been investigated functionally to elucidate a biologically plausible contribution to pathogenesis. Even fewer mutations have been studied in a genetically engineered
animal model or native cardiac cell. Computational strategies have been developed to predict the functional consequences of mutations, but none of these methods have been tested rigorously as valid clinical predictors. The lack of functional or biological validation of mutation effects remains the most severe limitation of genetic test interpretation in the cardiac channelopathies (88).

Interpretation of a negative genetic test result in a symptomatic person is a challenge. Despite more than 18 years of genetic discovery in the cardiac channelopathies, there remain a substantial number of cases having classic symptoms and signs for one of the heritable arrhythmia syndromes who test negative for the many known genes. This may be explained by a false-negative or true-negative test result. One potential cause for a false-negative genetic test result is the location of a mutation outside of the region of the gene normally interrogated by the test (92). Alternatively, certain types of mutation may be missed by standard testing strategies. For example, DNA sequencing can miss multi-exon deletion or duplication mutations (93–95). False-negative results may be overcome sometimes by repeat testing in situations where the clinical diagnosis has a high level of certainty (96). A true-negative test result may be a clue to the existence of an as-yet undefined gene involved with arrhythmia susceptibility. These situations are excellent opportunities to pursue genetic discovery in a research setting, as was recently done to identify novel calmodulin gene mutations in severe infantile cardiomyopathy (89,98). These and related observations have inspired the hypothesis that genetic factors other than the primary disease-associated mutation can modify the risk for disease-related morbidity and mortality. Conceptually, hypotheses proposed to explain variable penetrance in the genetic arrhythmias may be separated into 2 categories: 1) factors that modify the underlying arrhythmogenic myocardial substrate; and 2) factors that affect the probability and magnitude of arrhythmia-triggering events. Genetic factors that could affect the myocardial substrate include genes that encode proteins that contribute to the balance of inward and outward currents operating during the cardiac action potential. Genetic factors responsible for inter-individual differences in sympathetic and parasympathetic tone may alter one’s susceptibility to triggered arrhythmias. Likewise, the magnitude of catecholamine responses to stress and exercise varies among individuals, and some of this variability may have a genetic basis (100). Therefore, genes that participate in autonomic responses are candidate genetic modifiers.

Sorting out the relative effects of genetic modifiers is challenging. Demonstrating association of particular genetic variants with phenotype requires a large population, and there are sample size restrictions with any rare disease, such as inherited arrhythmias. Exploiting unique cohorts such as founder populations may have particular value in identifying modifier genes (97,101). For example, common variants in NOS1AP originally tagged in association with variable QT interval duration in healthy adults have been demonstrated to be modifiers of both the QT interval and the probability of symptoms in LQTS (102,103). Also, common variants in the 3’ untranslated region of KCNQ1 modify disease severity in an allele-specific manner (104).

Ideally, risk-stratification schemes based on the presence of a primary mutation and one or more modifier alleles will emerge to improve prediction of cardiac events. However, there are challenges to extrapolating from population-based results to predicting an individual’s risk. Clearly, more work is needed before we can take full advantage of this information for the ultimate goal of assessing risk even in asymptomatic mutation carriers.

Genetic modifiers. Another important conceptual barrier to extrapolating genetic test results to patient management is the variable disease expression and penetrance common among these disorders. For example, in congenital LQTS, not all individuals carrying disease-associated mutations have equal risk for expressing the clinical manifestations of the disease (34,97). Clinical heterogeneity is a common feature in LQTS and BrS. Members of the same family who share the same mutation may have varying phenotypes, ranging from no symptoms to sudden death. In rare cases, multiple mutations or combinations of a mutation with a common variant have accounted for unusual severity of one member of a larger family. Compound mutations help explain exaggerated disease severity in 4% to 8% of LQTS probands (98,99). On the other hand, predicting the likelihood of life-threatening arrhythmias in an asymptomatic mutation carrier continues to be most challenging.

Genetics and medico-legal implications. The effectiveness of cascade screening for the early identification of affected family members also carries medico-legal implications. Cascade screening requires positive genotyping of the proband because identification of the disease-causing mutation is the necessary first step. It follows that the physician who does not attempt to genotype the proband clinically affected by one channelopathy has willfully decided to ignore whether some of the family members are carriers of the disease and thereby are exposed to the risk of life-threatening arrhythmias. Likewise, the physician who, after having obtained positive genotyping, does not propose to initiate cascade screening within the family of the proband has similarly willfully decided to leave the affected family members—approximately one half of the first-degree relatives—informed about their status and unprotected.
Future impact of genetics. Advances in DNA sequencing technology have inspired a vision of widespread use of genome sequencing in clinical medicine. Exactly how this vision will be realized is uncertain at the present time, but there is enormous potential for affecting risk prediction for common and less common diseases and for predicting responses to drug therapy, including drug-induced TdP, which can be favored by specific genetic variants (105,106). Whole genome sequencing will eventually achieve an accuracy level and price point that will supplant targeted genetic testing for rare diseases, such as inherited arrhythmia syndromes. Although this may make diagnosing genetic disorders technically more feasible, the art of making a clinical diagnosis will remain important particularly when genetic information reveals unexpected findings. There is already information from large-scale exome sequencing efforts to anticipate that many incidentally discovered genetic variants in disease-associated genes, including those associated with LQTS, will be discovered in many individuals (107). Many of these variants could be merely false-positive results, and strategies to properly interpret and handle these incidental findings will be critical to avoid evoking needless concern or implementing unnecessary therapies in an asymptomatic person.

Conclusions
The progress in understanding channelopathies and the underlying molecular biology proceeds at mind-boggling speed. It should be clear to everyone in cardiology and medicine that genetics and clinical management of these diseases are tied together and that nowadays it is seldom possible to efficiently treat the affected patients without taking into account what has been learned from genetic testing.

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