

Heritable arrhythmias associated with abnormal function of cardiac potassium channels

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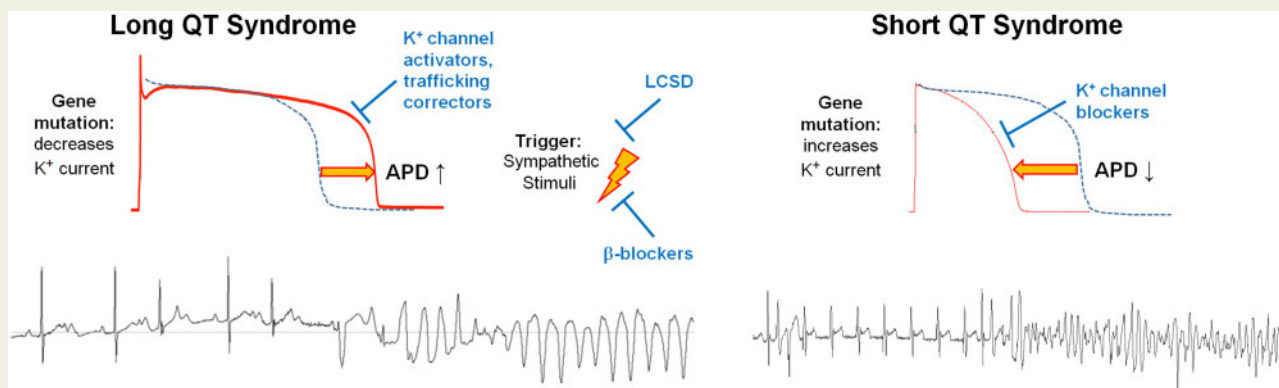
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Abstract

Cardiomyocytes express a surprisingly large number of potassium channel types. The primary physiological functions of the currents conducted by these channels are to maintain the resting membrane potential and mediate action potential repolarization under basal conditions and in response to changes in the concentrations of intracellular sodium, calcium, and ATP/ADP. Here, we review the diversity and functional roles of cardiac potassium channels under normal conditions and how heritable mutations in the genes encoding these channels can lead to distinct arrhythmias. We briefly review atrial fibrillation and J-wave syndromes. For long and short QT syndromes, we describe their genetic basis, clinical manifestation, risk stratification, traditional and novel therapeutic approaches, as well as insights into disease mechanisms provided by animal and cellular models.

Graphical Abstract



Keywords

Long QT syndrome • Short QT syndrome • Potassium channels • Atrial fibrillation • Sudden cardiac death

This article is part of the Spotlight Issue on Inherited Conditions of Arrhythmia.

1. Potassium currents in the heart: functional roles in repolarization and arrhythmias

1.1 A plethora of K^+ channels in the heart

Cardiomyocytes express a wide variety of K^+ channel proteins that conduct at least 11 different types of K^+ selective currents. This diversity includes three delayed rectifier currents (I_{Kr} , I_{Ks} , and I_{Kur}), three inward rectifier currents (I_{K1} , I_{KATP} , and I_{KACh}), two transient outward currents (I_{tof} and I_{tos}), two intracellular cation-activated currents (I_{KNa} and I_{KCa}), and at least one 'background leak' current (I_{leak}). The primary physiological functions of these currents are to maintain the resting membrane potential (I_{K1}), mediate repolarization of action potentials (I_{tof} , I_{tos} , I_{Kur} , I_{Kr} , and I_{Ks}), or to promote repolarization in response to an elevation of $[Na^+]_i$ (I_{KNa}), $[Ca^{2+}]_i$ (I_{KCa}), or a reduction in the $[ATP]/[ADP]_i$ ratio (I_{KATP}). In the past few decades, the molecular identity of the channels that conduct these currents have been identified, and the specific contribution of each channel type to cardiac excitability and mechanisms of their modulation by accessory subunits and intracellular messengers have been defined. The voltage-gated K^+ (Kv) or ligand-gated channels that conduct I_{tof} , I_{tos} , I_{Kur} , I_{Kr} , I_{Ks} , I_{KNa} , and I_{KCa} function as tetrameric complexes that are formed by coassembly of four identical or highly similar α -subunits that each contain six transmembrane segments (S1–S6) that constitute two distinct functional domains. The S1–S4 segments form a voltage-sensing domain, and the S5–S6 segments from each subunit surround the central cavity of the channel and provide the transmembrane conduit for selective K^+ flux. Inwardly rectifying K^+ (Kir) channels that conduct I_{K1} , I_{KATP} , and I_{KACh} are formed by coassembly of four smaller subunits that each have two transmembrane segments equivalent to the pore-forming S5–S6 of Kv channels. In addition, functional Kv and some Kir channels also include accessory (β) subunits that modulate gating and/or trafficking of channels to the cell membrane. The relative magnitudes of each K^+ current varies as a function of both time and voltage during a cardiac action potential and is schematically illustrated in Figure 1 for a human atrial and ventricular myocyte.

1.2 Molecular identity and physiological roles of delayed rectifier K^+ channels

Cardiac delayed rectifier K^+ currents are distinguished from one another based on their relative rates of activation onset: I_{Ks} is slow, I_{Kr} is rapid, and I_{Kur} is ultra-rapid. I_{Ks} is conducted by Kv7.1 (*KCNQ1*) channels² that are coupled to a variable number of minimal K^+ (minK) β -subunits (*KCNE1*) that each have a single transmembrane domain (gene names are indicated in italics). Kv7.1 channels activate fast in response to membrane depolarization and also inactivate at potentials >0 mV. In contrast, I_{Ks} conducted by Kv7.1/minK channels activate much more slowly and do not inactivate³ and thus, outward current increases throughout the plateau phase of the action potential and reaches its peak amplitude during the onset of Phase 3 repolarization. I_{Kr} is conducted by channels formed by coassembly of four Kv11.1 subunits (*KCNH2* or *HERG1*)^{4,5} and is characterized by a rapid rate of activation (relative to I_{Ks}) and even faster rate of inactivation, a property that greatly limits the magnitude of outward current during Phase 2 (plateau) of the action potential. Kv11.1 channels recover from their non-conducting, inactivated state to a conducting (open) state during Phase 3 repolarization, resulting in an enhanced current despite the reduced electrochemical driving force for K^+ . In the mammalian heart, I_{Kr} channels include full-length subunits

(Kv11.1a) that coassemble with a smaller, alternatively spliced variant (Kv11.1b) that accelerate the rate of channel closure.⁶ I_{Kur} is restricted to the atria in the human heart⁷ and is conducted by Kv1.5 (*KCNA5*) channels that activate with extremely fast kinetics and can also exhibit voltage-dependent inactivation when associated with ancillary subunits (Kv β 1.2, Kv β 1.3, or Kv β 2.1) that alter gating kinetics and modulate channel trafficking to the plasma membrane.⁸

1.3 Molecular identity and physiological roles of transient outward K^+ channels

Cardiac transient outward K^+ current (I_{to}) is very rapidly activated during the upstroke of the action potential and then more slowly inactivates. The kinetic properties of I_{to} promote the initial and short-lived period of repolarization (Phase 1) that precedes and modulates the duration and amplitude of the plateau (Phase 2) of action potentials (Figure 1). Two types of I_{to} are distinguished in the heart based on their relative rates of recovery from inactivation.⁹ I_{tof} recovers fast and is conducted by channels composed by coassembly of Kv4.3 (*KCND3*) and/or Kv4.2 (*KCND2*) α -subunits. I_{tos} recovers slowly from inactivation and is conducted by Kv1.4 (*KCNA4*) channels. In the human heart, Kv4.3 is coassembled with KChIP2 (*KCNIP2*) ancillary subunits that modulate channel gating and control the trafficking of channels to the cell surface.^{10,11}

1.4 Molecular identity and physiological roles of inward rectifier K^+ channels

Kir channels exhibit marked inward rectification, meaning that inward currents activated at potentials negative to the K^+ equilibrium potential (E_K) increase as a linear function of voltage, whereas outward currents activated at potentials positive to E_K exhibit a progressively reduced slope conductance that culminates in very little sustained current during the plateau phase of a cardiac action potential. Reduced outward current at positive potentials is caused by voltage-dependent block of Kir channels by intracellular polyamines and Mg^{2+} .¹² All Kir channels are constitutively activated by phosphatidylinositol-4,5-bisphosphate (PIP_2).¹³ Cardiac I_{K1} is mediated by Kir2 channels, mainly Kir2.1 (*KCNJ2*) but also Kir2.2 (*KCNJ12*) and Kir2.3 (*KCNJ4*) that are constitutively open and thus, are largely responsible for setting the resting membrane potential of atrial and ventricular myocytes to a potential close to E_K .¹⁴ I_{K1} is smaller in atrial than in ventricular myocytes and is relatively absent in myocytes in the sinoatrial node (SAN) and atrioventricular node that consequently have a less negative maximal diastolic potential. I_{KACh} is restricted to the atrium and is conducted by channels formed by Kir3.1 (*KCNJ3*) and Kir3.4 (*KCNJ5*) α -subunits.¹⁵ I_{KACh} is activated by extracellular binding of acetylcholine to M2 muscarinic receptors and results in shortening of action potential duration (APD) in atrial myocytes and a hyperpolarization and slowing of the firing rate of SAN cells. I_{KATP} channels are hetero-octomeric complexes, formed by coassembly of four Kir6.2 (*KCNJ11*) and/or Kir6.1 (*KCNJ8*) α -subunits plus four SUR2A or SUR1 accessory subunits.¹⁶ K_{ATP} channels are activated by intracellular MgADP, inhibited by intracellular ATP ($IC_{50} \sim 50 \mu M$), and regulation by the ATP/ADP ratio is modified by channel phosphorylation and PIP_2 .¹⁷ K_{ATP} channels are normally in a closed state but are readily activated and can dramatically shorten APD during conditions that reduce $[ATP]_i$ such as hypoxia or ischaemia.

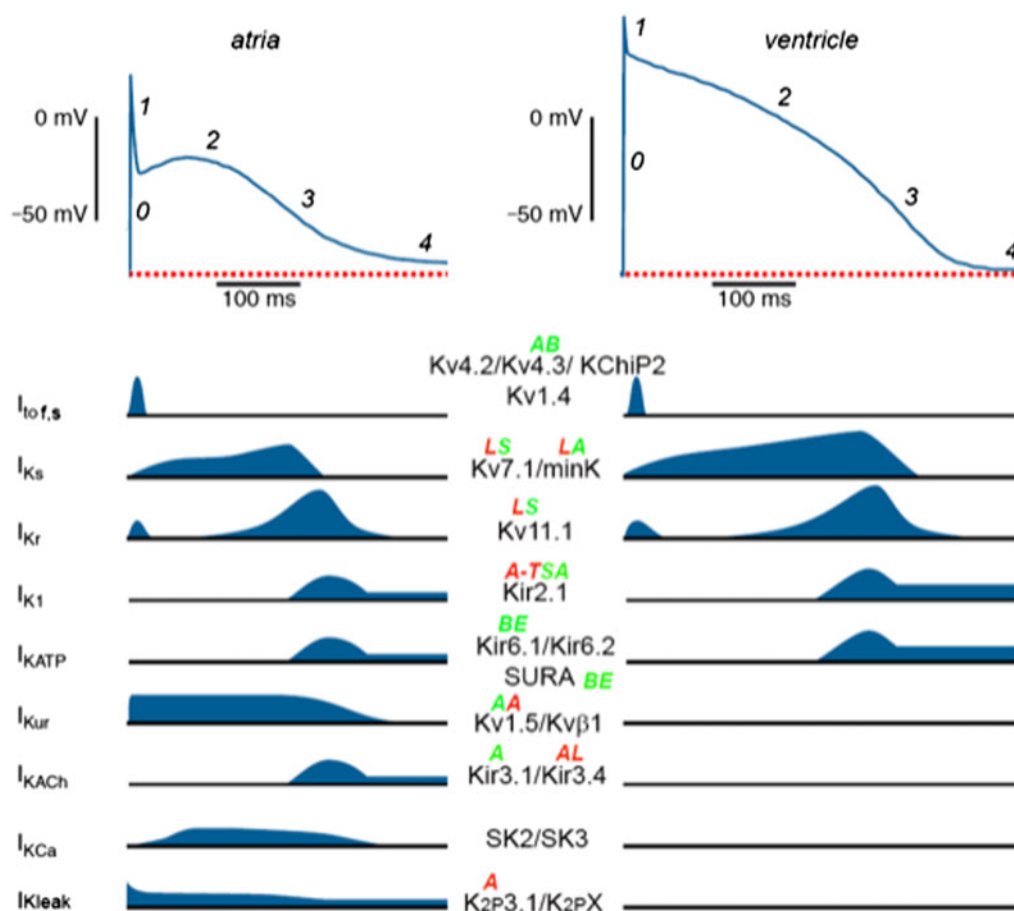


Figure 1 Multiple potassium ion currents mediate repolarization of human cardiomyocytes. Upper panels indicate the timing of the different phases 0–4 of a typical atrial (left) and ventricular (right) action potential. Lower panel shows approximate amplitude of each current throughout the duration of the action potentials. Current name is indicated at the far left. The primary K^+ channel subunits that form channels that conduct these currents is indicated in the centre (in black text); cardiac arrhythmia known to be associated with a specific subunit is also indicated according to the code: **A**, atrial fibrillation; **A–T**, Anderson–Tawil syndrome; **B**, Brugada syndrome; **E**, early repolarization syndrome; **L**, LQTS; **S**, SQTs; Colour of text indicates type of mutation, either a gain of function (green) or loss of function (red). Note that current magnitude can vary significantly under different conditions; for example, I_{KATP} and I_{KNa} (not shown) can range from unmeasurable under normal physiological conditions to very large during anoxia. Modified with permission from Schmitt et al.¹

1.5 Molecular identity and physiological roles of cation-activated and leak K^+ channels

Cardiac myocytes also express channels that conduct intracellular cation-activated currents (I_{KCa} , I_{KNa}) and a background 'leak' current (I_{kleak}). Large conductance Ca^{2+} -activated (BK) channels are located in mitochondrial membranes, but sarcolemma I_{KCa} is conducted by small conductance Ca^{2+} -activated K^+ (SK) channels SK1–SK3 (*KCNN1–3*)¹⁸ that are expressed primarily in atrial myocytes where they are activated in response to elevated $[Ca^{2+}]_i$ subsequent to the opening of sarcolemma L-type and sarcoplasmic reticulum RyR Ca^{2+} channels. Ca^{2+} activates SK channels by binding to calmodulin that constitutively interacts with the C-termini of SK α -subunits.¹⁹ In human atria, SK2 and SK3 expression levels are ~10-fold higher than that of SK1, and electrical remodelling associated with chronic atrial fibrillation (AF) reduces their expression by ~50%.²⁰ I_{KNa} is conducted by K_{Na} 1.2 (*KCNT2*) channels that are expressed in the sarcolemma and perhaps mitochondrial

membranes of cardiomyocytes.²¹ Although K_{Na} 1.2 channels have a very high conductance, the outward current conducted by these channels is predicted to be quite small except under severe ischaemia when $[Na^+]_i$ can increase significantly. Opening of these channels in the mitochondria has been proposed to counteract opening of the mitochondrial permeability transition pore and thus protect against ischaemia–reperfusion injury.²² I_{kleak} is the term used here to describe the prominent background K^+ current in atrial myocytes. It is likely that several two-pore domain K^+ (K_2P) channels that are voltage independent and gated by pH, lipids and membrane stretch contribute to this current. In the human heart, K_2P 3.1 (TASK1, *KCNK3*) channels were initially reported to be expressed predominantly in the atria,²³ where they can form as homodimers or heterodimers when partnered with K_2P 9.1 (TASK-3, *KCNK9*).²⁴ More recently, it was reported that at least 10 different K_2P channels are expressed, albeit at low levels, in the human ventricle.²⁵ The specific role(s) of these channels in ventricular repolarization is unclear, but APD prolongation was noted after RNAi-mediated knock-down of K_2P 2.1, K_2P 3.1, K_2P 6.1, and K_2P 17.1 expression in human iPSCs

differentiated into cardiomyocytes.²⁵ A role for K2P2.1 in arrhythmogenesis is also suggested by the finding that mRNA and protein levels are down-regulated in heart failure and AF.²⁶

Ion channels are regulated by a plethora of molecular interactions with accessory subunits, kinases, chaperones, signalling networks, etc. that affect their levels of expression, intracellular trafficking, rates of turnover, and single channel gating properties such as open probability and rates of activation and inactivation. Most cardiac K⁺ channels are modulated by phosphorylation by one or more enzymes such as PKA, PKC, CaMKII, or Src.¹ As one example, *I_{Ks}* channels are bound to the A kinase-anchoring protein AKAP9,²⁷ which in turn recruits the binding of PKA, protein phosphatase 1 (PP1), phosphodiesterase 4D3,²⁸ and adenylyl cyclase 9 to the channel complex.²⁹ *I_{Ks}* channels also interact with calmodulin, PIP₂ and ATP that stabilize the channel in an open state.^{30–32} For in depth descriptions of the myriad ways in which cardiac K⁺ channels can be regulated, the reader is directed to several extensive review articles on this topic.^{1,33,34}

The discovery of mutations in genes that encode many K⁺ channel subunits has provided unique insights into the molecular mechanisms and pathophysiology associated with genetic forms of AF and ventricular fibrillation (VF). In general terms, loss of function mutations in K⁺ channel subunits results in a delayed time course of cardiac repolarization (e.g. long QT syndrome, LQTS), whereas gain of function mutations results in a faster rate of cardiomyocyte repolarization (e.g. AF and short QT syndrome, SQTs). Heterozygous missense mutations are the most common cause of K⁺ channelopathies and most of these single amino acid substitutions cause a loss of normal function, either by causing protein misfolding and early degradation, hindering trafficking of the channel to the cell membrane or by altering gating in a manner that reduces net conductance (e.g. altered kinetics or voltage dependence of activation or inactivation). Below we present a brief consideration of AF and J-wave syndromes, followed by a more in-depth discussion of LQTS and SQTs. Finally, it should be noted that K⁺ channel mutations can also cause a wide spectrum of human diseases beyond cardiac arrhythmia. Notable examples are epilepsy,³⁵ diabetes,³⁶ ataxia,³⁷ and Cantu syndrome that includes atrioventricular (AV) node block.³⁸

2. Atrial fibrillation and J-wave syndromes

2.1 Atrial fibrillation

Several K⁺ channels have been reported to be associated with rare forms of genetic AF, including Kv1.5, Kv4.2, Kv4.3, Kir2.1, Kir3.4, and K_{2P}3.1. The first K⁺ channel associated with AF was Kir2.1 (*KCNJ2*) where a gain of function mutation (V93I) was detected in all affected members of a Chinese kindred.³⁹ The following year, the first report for Kv1.5 (*KCNA5*) was a heterozygous E375X mutation predicted to result in a loss of function by introducing a premature stop codon that eliminates the S4-terminus region of the channel subunit.⁴⁰ Early-onset nocturnal paroxysmal AF in a family was found to be caused by a point mutation (S447R) that resides in a PKC phosphorylation site of Kv4.2 (*KCND2*) channels.⁴¹ This mutation increased *I_{to}* magnitude by slowing inactivation and enhancing membrane expression of channels. A point mutation (A545P) in Kv4.3 (*KCND3*) increases peak-current density and slows the onset of inactivation of *I_{to}*.⁴² A role for *I_{KACH}* in AF has been suggested by the initial finding that common polymorphisms in Kir3.4 (*KCNJ5*) are associated with lone AF,⁴³ and more recently by the report

of five rare mutations in both Kir3.1 (*KCNJ3*) and Kir3.4 (*KCNJ5*) in patients with sporadic AF.⁴⁴ One gain of function mutation (N83H) in Kir3.1 enhances constitutively active *I_{KACH}* (M2 receptor stimulation not required),⁴⁴ a cellular phenotype noted previously in atrial myocytes isolated from AF patients.⁴⁵ Most recently, a gain of function mutation in Kir3.4 was reported to cause sinus node dysfunction in a large family.⁴⁶ Point mutations in Kv β -subunits minK related protein 2, MiRP2 (*KCNE3*),⁴⁷ and MiRP4 (*KCNE5*)⁴⁸ have also been reported in single patients with AF. KCNE subunits are especially interesting because they modulate the gating of several different Kv channels,⁴⁹ a feature that also makes it difficult to define the mechanisms underlying their pathology in AF. Loss of function variants in K_{2P}3.1 has also been associated with AF in a few patients.⁵⁰

2.2 J-wave syndromes

The term J-wave syndromes was recently suggested to describe ventricular arrhythmia associated with both Brugada syndrome and early repolarization syndrome (ERS) that are characterized by VF and elevation and notching of the J-point (a deflection immediately following the QRS complex) on the electrocardiogram (ECG).⁵¹ These arrhythmias are very rarely related to an enhanced outward *I_{to}* or *I_{KATP}* resulting from gain of function mutations in Kv4.3 (*KCND3*)⁵² or Kir6.1 (*KCNJ8*),⁵³ or loss of function mutations in the Kv4.3 β -subunit MiRP2 (*KCNE3*).⁵⁴ Most recently, a *de novo* mutation in Kv4.3 (G306A) was reported from an ERS-associated proband and shown to increase the magnitude of *I_{to}* and slow the rate of its onset and recovery from inactivation.⁵⁵

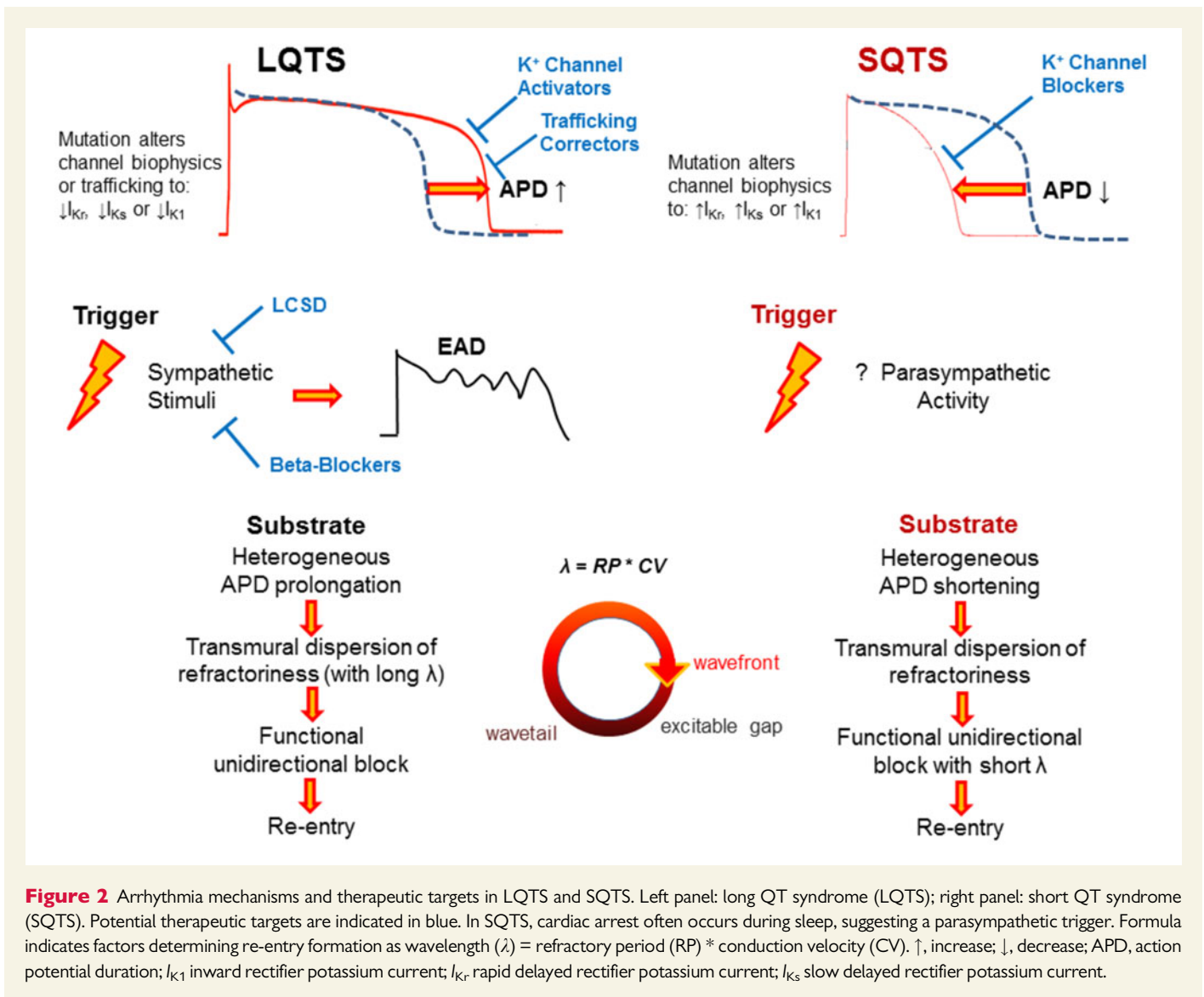
3. Long QT syndrome

LQTS is a genetically transmitted cardiac disease characterized by prolonged QT interval of the surface ECG, caused by a prolonged duration in the plateau phase of ventricular action potentials.⁵⁶ Prolonged repolarization can result from a reduction of outward currents, particularly *I_{Kr}* and *I_{Ks}* or by an enhancement of inward Na⁺ or Ca²⁺ currents during Phase 2 or 3 of the action potential. Patients affected by LQTS have a structural normal heart, but are at high risk of life-threatening arrhythmias, because the prolongation of repolarization may result in subsequent re-activation of Ca²⁺ or Na⁺ currents, which in turn may generate an early after depolarization (EAD) and promote triggered electrical activity at the end of repolarization⁵⁶ (Figure 2). The typical arrhythmia in LQTS patients is Torsades de Pointes (TdP), a tachyarrhythmia that can self-terminate or degenerate into VF.⁵⁶ Clinically, the patients can suffer palpitations, syncopal events or sudden cardiac death (SCD), usually starting from childhood or teen age. An appropriate diagnosis is mandatory as effective therapies are available and prevention of SCD is possible.⁵⁶

3.1 Molecular mechanisms

Two main hereditary variants of LQTS are known; the autosomal dominant Romano–Ward syndrome,⁵⁷ with a prevalence of 1:2000 live births⁵⁸ and the recessive Jervell–Lange Nielsen (JLN) syndrome, in which a more severe cardiac phenotype is associated with congenital deafness.⁵⁹

Seventeen genes are currently associated with LQTS (LQT1–LQT17) and among these, seven encode for K⁺ channel subunits or for proteins that modulate K⁺ channel function. In absolute numbers, mutations in K⁺ channel genes are responsible for the vast majority of LQTS cases. Specifically, *KCNQ1* and *KCNH2* encoding Kv7.1 and Kv11.1 α -subunits



of I_{Ks} and I_{Kr} channels are responsible for LQT1 and LQT2. Together, mutations in these genes account for 80% of all LQTS cases. Perturbation in I_{Ks} and I_{Kr} can also occur when β -subunit genes as *KCNE1* (LQT5 or JLN) and *KCNE2* (LQT6) are mutated. These are responsible for less than 5% of all LQTS cases^{60,61} and LQT5 is usually associated with a low penetrant form of the disease.⁶² Other rare genetic subtypes characterized by a reduction in outward K^+ current are LQT7 or Andersen–Tawil syndrome (due to mutations in *KCNJ2* that reduce I_{K1}), LQT11 (due to mutation in *AKAP9*, the gene encoding for Yotiao, an I_{Ks} channel modulator), and LQT13 (due to mutation in *KCNJ5* that reduces I_{KACH}).⁵⁶

LQTS mutations can affect multiple properties of channel function (as reviewed in detail in Ref.⁶³) Mechanisms of reduced Kv7.1/minK or Kv11.1 channel function include protein misfolding, misprocessing in the endoplasmic reticulum, disrupted trafficking of channels to the cell surface, reduced K^+ permeation, and altered channel gating, such as enhanced inactivation, slower activation or faster deactivation. A single virulent mutation in only one subunit of a tetrameric Kv and Kir channel can sometimes have a ‘poison pill’ effect and result in complete

dominant-negative suppression of channel function. For example, mis-sense mutations in the intracellular C-helix of Kv11.1 that interrupts its interaction with minK subunits decreases I_{Ks} density and also impairs PIP₂ modulation, causing a depolarizing shift of channel activation.⁶⁴

3.2 Clinical presentation

The clinical presentation of LQTS varies significantly in different patients—ranging from no symptoms to sudden death. Arrhythmic events are usually triggered by an increase in sympathetic activity and therefore, the most typical presentation is a syncopal event during emotional or physical stress in a young and otherwise healthy individual. In LQT1 subjects, cardiac events typically occur during exercise, with swimming being a specific trigger in one-third of the cases.⁶⁵ Conversely, in LQT2, the predominant trigger is emotional stress, with sudden auditory stimuli being pathognomonic for this genetic subgroup.⁶⁵ LQT2 patients are also more susceptible to QT prolonging drugs and electrolyte unbalances and female patients are at higher risk in the post-partum period⁵⁶ and during menopause transition and post-menopausal periods.⁶⁶

Table 1 Long QT syndrome diagnostic criteria

Points		Points	
Electrocardiographic findings^a		Clinical history	
QTc ^b		Syncope ^c with stress	2
≥480 ms	3	Syncope ^c without stress	1
460–479 ms	2	Congenital deafness	0.5
450–459 ms (in males)	1		
QTc 4th min of recovery from exercise stress test ≥480 ms	1	Family history	
Torsade de pointes ^c	2	Family members with definite LQTS ^d	1
T-wave alternans	1	Unexplained SCD below age 30 among immediate family members ^d	0.5
Notched T wave in three leads	1		
Low heart rate for age ^e	0.5	SCORE ≤1 point: low probability of LQTS	
		SCORE 1.5–3 points: intermediate probability of LQTS	
		SCORE ≥3.5 points high probability of LQTS	

Adapted from Schwartz and Crotti.⁶⁷^aIn the absence of medications or disorders known to affect these electrocardiographic features.^bQTc calculated by Bazett's formula.^cMutually exclusive.^dThe same family member cannot be counted in both.^eResting heart rate below the 2nd percentile for age.

Basal ECG, exercise stress test and 12-lead 24-h Holter monitoring are all extremely important to identify the main electrocardiographic features of the disease. Typical features on surface ECG include QTc prolongation (>440 ms for men and >460 ms for women after puberty) and alteration of T-wave morphology that can be biphasic, bifid or notched.⁵⁶

Typically, LQT1 patients tend to have broad-based T waves; while biphasic or notched T waves are more frequently observed in LQT2 patients. LQTS-related ECG features associated with major electrical instability include T-wave alternans and functional 2:1 AV block. T-wave alternans is a beat-to-beat alternation of the polarity and/or amplitude of the T wave,⁵⁶ while the 2:1 functional 'pseudo' AV block is usually seen in neonates or young children who have a particularly prolonged QT, leading to conduction block within the refractory ventricle.

In case of borderline QTc, it is very important to evaluate the QT behaviour during an exercise stress test and 24 Holter recording to make a diagnosis. Indeed, both the inability of QT to shorten during exercise and QT prolongation during the recovery phase have been identified as typical features of affected patients, particularly with LQT1.⁶⁷ Specifically, the presence of a QTc ≥480 ms at the fourth minute of recovery is one of the diagnostic criteria of the disease.⁶⁷ Regarding ECG Holter recording, phases of clear QT prolongation and variability in the morphology of the T wave could be highly suggestive or diagnostic for the disease. Furthermore, 24-h Holter recording may provide prognostic information; i.e. identification of T-wave alternans, pronounced QT prolongation, or frequent sudden pauses with abnormal repolarization abnormalities in the subsequent beat are all negative prognostic factors. Echocardiography in LQTS patients never show major structural abnormalities; however, subtle mechanical abnormalities do exist and correlate with arrhythmic risk.^{68–71}

Among potassium channel-related LQTS, there are two syndromic forms with peculiar features. The JLN syndrome is a severe variant of LQTS, in which the cardiac phenotype is combined with congenital deafness⁷² due to the presence of homozygous or compound heterozygous mutations in *KCNQ1* (LQT1) or *KCNE1* (LQTS) genes.⁵⁹ Patients with JLN syndrome present with a malignant phenotype: 90% of them are symptomatic (often already at paediatric age), the QTc is markedly prolonged (on average 557 ± 65 ms) and conventional therapies have limited

efficacy, leading to the recommendation of early implantable cardioverter-defibrillator (ICD) implantation, even in children. JLN subgroups at lower risk are those with a QTc <500 ms, those asymptomatic during the first year of life and those with *KCNE1* mutations.⁵⁹

The Andersen–Tawil syndrome (ATS), also referred to as LQT7 (mutations in *KCNJ2*), is characterized by prominent U waves on the ECG that are often difficult to distinguish from truly prolonged QT, ventricular arrhythmias and potassium-sensitive periodic paralysis. Affected patients may also present mild or prominent dysmorphic facial and limb features.^{73,74} Ventricular arrhythmias range from premature ventricular beats to short runs of polymorphic ventricular tachycardia (VT) and no specific trigger has been identified.^{73,74} It is quite debated if ATS should be considered as LQTS variant, as a true QT prolongation is frequently not present and arrhythmogenesis might be related to a different mechanisms than conventional LQTS.⁷⁵ Epilepsy is another extra-cardiac manifestation that can be observed in some LQTS cases⁷⁶; specific examples include patients with severe heterozygous loss of function mutations in *KCNH2*⁷⁷ or *KCNQ1*.⁷⁸

3.3 Diagnosis and risk stratification

The diagnosis of LQTS can be made if a Schwartz score (based on electrocardiographic features, familial and personal history⁶⁷) of ≥3.5 points is found (Table 1), or in presence of a QTc ≥500 ms in repeated 12-lead ECGs.⁷⁹ These criteria do not allow the identification of silent mutation carriers. Therefore, an LQTS diagnosis can also be made in presence of an unequivocally pathogenic mutation.⁷⁹

The main differential diagnosis is the so-called acquired long QT syndrome (aLQTS), in which QT prolongation is caused either by drugs, bradycardia or hypokalaemia. Also in aLQTS a genetic predisposition can be identified, and K⁺ channel genes play a predominant role.⁸⁰

Risk stratification in LQTS is based on clinical and genetic factors.⁸¹ Patients symptomatic for cardiac events (syncope or cardiac arrest) are unquestionably at higher risk of recurrences if not adequately treated,⁸² with those having events in the first year of life being the most severe cases.⁸³ Another major risk factor is a QTc >500 ms,⁸⁴ with additional malignant ECG features being T-wave alternans, 2:1 AV block and sinus pauses followed by particularly prolonged QT intervals. In the LQT1

subgroup, autonomic parameters have been identified in a few studies as useful in risk stratification, such as reduced baroreflex sensitivity or reduced heart rate reduction during the first minute of recovery from an exercise stress test.^{85,86} Carrying two independent mutations⁸⁷ or having JLN syndrome^{59,72} is associated with an increased arrhythmic risk, but even in the presence of a single disease-causing mutation the risk can be influenced by the gene involved,⁸⁴ the location of the mutation in the protein,^{88,89} and sometimes by the specific mutation.⁹⁰

As in many inherited disorders, there is considerable variability of disease expressivity in LQTS; i.e. even within members of the same family carrying the same disease-causing mutation, clinical manifestation can be very different. Such an interpersonal variation is partially attributed to genetic modifying factors, such as single-nucleotide polymorphisms (SNPs) that may attenuate or aggravate the pathological manifestation of the principal disease-causing mutation, thereby conferring protection or increased risk, respectively.⁹¹ A large number of modifier SNPs have been described since 2005,⁹² most of them affecting K⁺ channel function/expression^{92–95}; however, SNP evaluation is not yet integrated into clinical practice. Among different modifier SNPs so far described, those more strongly and reproducibly associated with arrhythmic risk in multiple studies^{96–98} are those on *NOS1AP*, a gene encoding for CAPON, an anchoring protein for the neuronal isoform of NO-synthase that is abundantly expressed in cardiac myocytes and highly relevant to intracellular Ca²⁺ handling.⁹⁹

Apart from the genetic modifiers other factors so far identified as possible modulator of disease severity are medications that could influence QT interval (www.crediblemeds.org) and therefore arrhythmic risk, alterations in serum potassium and magnesium levels, autonomic nervous system variations,^{85,86} and hormonal changes.^{56,66}

3.4 Therapy

The first line therapy for LQTS is β -adrenergic receptor blockers that should be started upon initial diagnosis as SCD can be the first manifestation of the disease in 12% of untreated patients.⁵⁶ Not all β -blockers are equally effective and nadolol and propranolol should be considered the β -blockers of choice, being associated with a lower risk of recurrences compared to metoprolol.¹⁰⁰ However, in a recent study, nadolol appeared to be more effective than propranolol, and this could be due to the better compliance obtained with nadolol (requiring only once a day administration instead of three times) and the fact that propranolol is particularly used in very small children at higher risk.⁸¹ In LQT3 patients with a QTc >500 ms it is recognized by current guidelines that mexiletine, a Na⁺ channel blocker, should be added to β -blocker therapy whenever a QT shortening of at least 40 ms is observed during an acute oral test.⁷⁹ Interestingly, a recent study also reported a significant QT shortening by mexiletine in LQT2 patients, suggesting a possible anti-arrhythmic effect also in this genetic subgroup.¹⁰¹

Whenever patients remain at high risk despite a full dose of β -blockers, or when β -blockers are not tolerated, left-cardiac sympathetic denervation (LCSD) should be considered. LCSD is a surgical procedure, that can also be performed with a thoracoscopic approach, that requires the removal of the first four thoracic ganglia (T1–T4), leaving intact the cephalic portion of the left stellate ganglion to avoid the Horner's syndrome.⁵⁶ The rationale for LCSD is mainly based on its antifibrillatory effect and on the major reduction of norepinephrine release without interfering with heart rate.¹⁰² In a cohort of high-risk LQTS patients that underwent LCSD, the procedure showed a 91% reduction in cardiac events and produced a mean QTc shortening of 39 ms, pointing to an action on the arrhythmic substrate as well as on its trigger.¹⁰³ Indeed,

LCSD reduces the inhomogeneous norepinephrine release in the ventricle with a consequent reduction of the dispersion of repolarization and therefore of the QTc.¹⁰⁴ In LQTS, an ICD is needed in a minority of the patients; those with a previous cardiac arrest and with recurrences despite full antiadrenergic therapy (possibly including LCSD).¹⁰⁵ Asymptomatic patients should not be implanted with an ICD, unless despite optimal therapy a QTc >550 ms is present together with signs of high electrical instability (i.e. T-wave alternans, 2:1 AV block, very long sinus pauses with subsequent beats showing aberrant T-wave morphology and further QT prolongation).¹⁰⁵

3.5 Cellular and animal models

Animal models for K⁺ channelopathies are very useful to investigate pathophysiological mechanisms of arrhythmogenesis on multiple levels (from cellular/tissue to whole heart and animal), to test the role of genetic background in altering the arrhythmic phenotype and to test novel therapies. However, the major limitation of animal studies is the partially limited clinical translation due to some species differences in cardiac electrical function.¹⁰⁶ Transgenic mouse models representing LQT1 or LQT2 (with knock-out or dominant-negative loss-of-function mutations of human voltage-gated K⁺ channels *KCNQ1* or *KCNH2*) have failed to mimic the human disease phenotype, particularly lacking spontaneous VT, VF, and SCD. The main reason is due to difference in the predominant repolarizing voltage-gated K⁺ channel currents responsible for cardiac repolarization in murine and human cardiomyocytes.¹⁰⁷

Rabbits represent a species with more pronounced similarities to human cardiac electrophysiology, e.g. with *I_{Kr}* and to a lesser degree, *I_{Ks}* as main drivers of cardiac repolarization.^{106,108} Several different transgenic rabbit models for LQTS have been generated based on a dominant-negative overexpression of human mutated K⁺ channels; e.g. LQT1 (*KCNQ1-Y315S*), LQT2 (*KCNH2-G628S*),¹⁰⁹ and LQT5 (*KCNE1-G52R*).¹¹⁰ In LQT1 or LQT2 rabbit cardiomyocytes, the repolarizing currents *I_{Ks}* (LQT1) or *I_{Kr}* (LQT2) were completely eliminated, resulting in a prolongation of APD, ventricular refractoriness, and QT prolongation in both,^{109,111} and the development of spontaneous VT and SCD in LQT2.¹⁰⁹ (Figure 3). In transgenic LQT5 rabbit cardiomyocytes, in contrast, the biophysical properties of *I_{Ks}* and *I_{Kr}* were altered with accelerated deactivation kinetics,¹¹⁰ but overall *I_{Ks}* and *I_{Kr}* current densities were not reduced. Consequently, these rabbits exhibited only a partial phenotype with no significant prolongation of whole heart APD and only a very slightly prolonged QT interval.¹¹⁰

Studies in transgenic LQT1 and LQT2 rabbits have strengthened our insights into the importance of spatial and temporal heterogeneities of repolarization as a pro-arrhythmic 'substrate' and the key role of increased sympathetic activity as a 'trigger' for EAD and LQTS-related arrhythmias. Indeed, in LQT2 rabbit hearts, a pronounced dispersion of repolarization was identified in left and right ventricles,^{109,111,113} causing unidirectional functional block and re-entry formation as the initiating mechanism of VT/VF.¹⁰⁹ In addition, in LQT2 hearts, a dynamic spatio-temporal dispersion of repolarization with discordant alternans in adjacent regions developed at physiological heart rates and preceded onset of VT/VF.¹¹⁴ In contrast, in LQT1 hearts with a more homogeneously prolonged APD within the left ventricle at normal heart rates,^{109,111,113,115} VT/VF did not develop spontaneously nor could it be induced, suggesting a protective effect of a homogeneous APD prolongation. When LQT1 hearts were further stressed, however, by continuous tachypacing to induce cardiac tachymyopathy,¹¹⁶ or complete AV block,¹¹⁷ APD dispersion increased, spatially discordant alternans developed and VT/VF was easily inducible.¹¹⁶

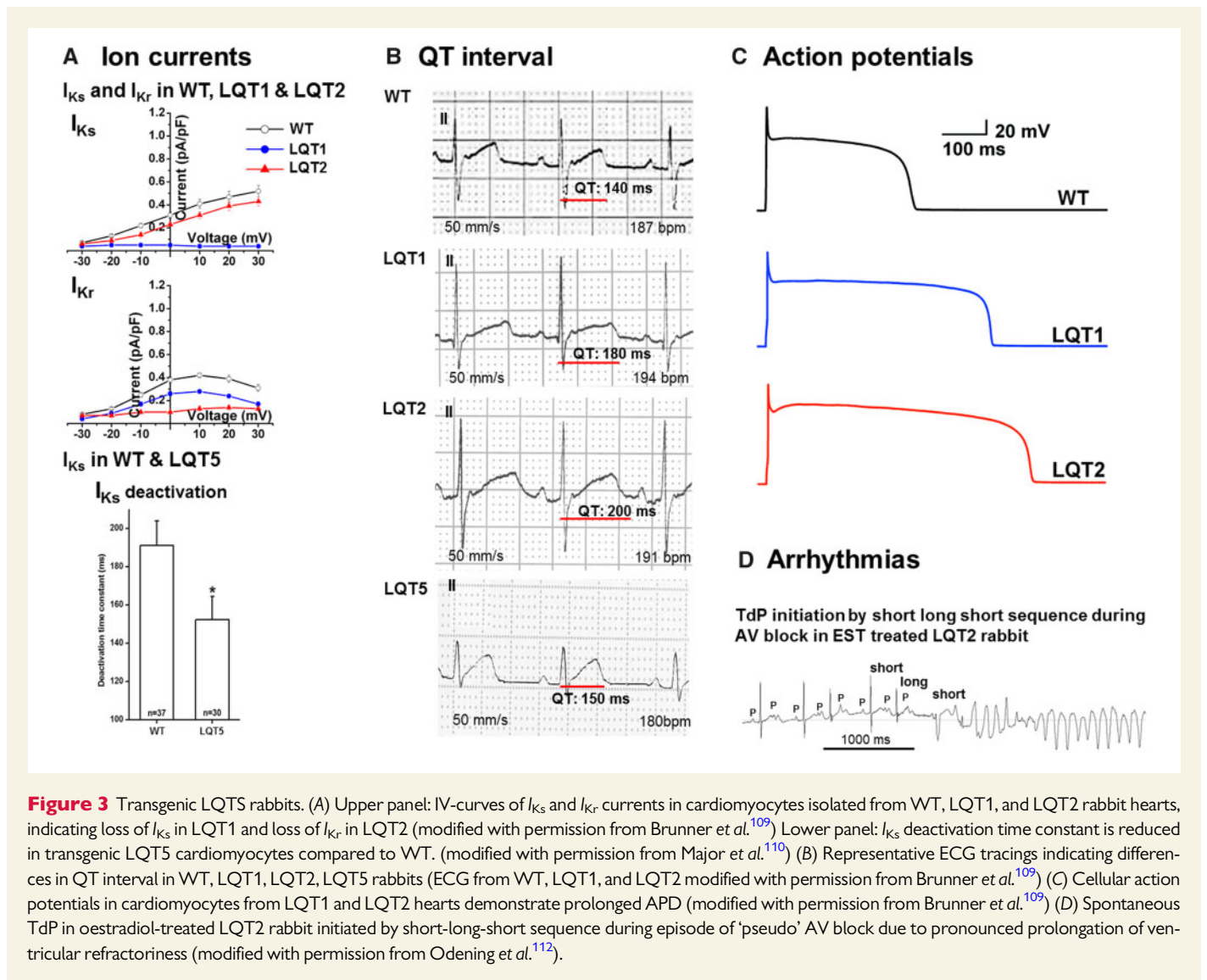


Figure 3 Transgenic LQTS rabbits. (A) Upper panel: IV-curves of I_{Ks} and I_{Kr} currents in cardiomyocytes isolated from WT, LQT1, and LQT2 rabbit hearts, indicating loss of I_{Ks} in LQT1 and loss of I_{Kr} in LQT2 (modified with permission from Brunner et al.¹⁰⁹) Lower panel: I_{Ks} deactivation time constant is reduced in transgenic LQT5 cardiomyocytes compared to WT. (modified with permission from Major et al.¹¹⁰) (B) Representative ECG tracings indicating differences in QT interval in WT, LQT1, LQT2, LQT5 rabbits (ECG from WT, LQT1, and LQT2 modified with permission from Brunner et al.¹⁰⁹) (C) Cellular action potentials in cardiomyocytes from LQT1 and LQT2 hearts demonstrate prolonged APD (modified with permission from Brunner et al.¹⁰⁹) (D) Spontaneous TdP in oestradiol-treated LQT2 rabbit initiated by short-long-short sequence during episode of 'pseudo' AV block due to pronounced prolongation of ventricular refractoriness (modified with permission from Odening et al.¹¹²).

Studies in transgenic LQT1 and LQT2 rabbit models have also been instrumental in evaluating the pro-arrhythmic, harmful, or anti-arrhythmic, beneficial effects of frequently utilized drugs,^{111,115,118} circulating metabolites,¹¹⁹ or hormones in LQTS.^{112,120} Moreover, several mechanisms underlying pro-arrhythmic effects of oestradiol and anti-arrhythmic, protective effects of progesterone have been identified in LQT2 rabbit models—the latter being based on shortening of repolarization, reduction of EAD formation, and Ca^{2+} stabilizing effects.¹¹² These studies suggest that progesterone-based therapies may be considered as novel anti-arrhythmic add-on approaches in female LQTS patients. Recently, potential harmful, pro-arrhythmic effects of postpartum hormones oxytocin and prolactin via I_{Ks} -inhibition have been revealed in the LQT2 rabbit models¹²⁰ that might contribute to the clinical observation of an increased postpartum arrhythmic risk - particularly in LQT2.⁹¹

In recent years, evidence has accumulated that in LQTS mechanical function is subclinically altered with impaired diastolic relaxation and prolonged contraction duration.^{68–70} Using phase-contrast-based magnetic resonance imaging, a regionally heterogeneous reduced diastolic function was demonstrated in transgenic LQT2 rabbits,¹¹³ thus

supplementing clinical findings with important insights into regional mechanical heterogeneity. In addition, a spatial correlation between the extent of electrical dysfunction (prolongation of APD) and impaired diastolic function¹¹³ and a correlation between the extent of mechanical dysfunction and arrhythmic risk was revealed.¹²¹ These experimental as well as first clinical data on differences in the extent of mechanical function among symptomatic and asymptomatic subjects⁷¹ suggest a potential future role for the assessment of mechanical dysfunction for risk stratification in LQTS. Moreover, the extent of mechanical dysfunction and its heterogeneity might not only reflect the extent of electrical dysfunction but may also be causatively linked to arrhythmia formation via mechano-electrical feedback.

Another animal model frequently used in long QT related research is the zebrafish.¹²² Zebrafish share pronounced similarities with human cardiac electrophysiology in terms of ventricular AP shape, AP/QT duration, and repolarizing ion currents—namely I_{Kr} .^{123,124} Furthermore, as the zebrafish is similarly sensitive to pharmacological modulations by I_{Kr} -blocking drugs, it has emerged as a model for drug safety testing.^{125,126} Rather than inducing LQTS by genetic manipulation of known disease causing mutations—as done in the rabbit and mouse LQTS models

highlighted in the previous sections—spontaneous LQTS zebrafish mutants caused by mutations in *zERG*, the zebrafish ortholog of *KCNH2*, do exist and can be identified in phenotype-based screening approaches.^{125,127} Among them, the zebrafish *breakdance* mutant, which carries the trafficking deficient *KCNH2-I59S* mutation, recapitulates several features of severe forms of human LQT2 including spontaneous EADs, and 2:1 AV block due to pronounced ventricular AP prolongation.^{127,128} Using this zebrafish model in a relatively high-throughput chemical screen has resulted in the identification of flurandrenolide and the novel compound 2-MMB, which are both able to rescue trafficking and may lead to novel therapeutic approaches of LQTS.¹²⁸ In addition to chemical screens for discovery and testing of novel drugs, zebrafish have also been used for phenotype-based screens to identify novel genes and gene networks implicated in the regulation of cardiac repolarization.¹²⁹

Animal models always show some species differences compared to human pathophysiology. Therefore, human diseases should ideally be studied in human organs, tissues, or cells. In recent years, induced pluripotent stem cell (iPSC) technologies have thus been utilized to investigate human iPSCs differentiated into cardiomyocytes (iPSC-CMs) derived from LQTS patients. Specifically, in 2010 and 2011, it was demonstrated in proof of principle studies that the three main genetic forms of LQTS can be modelled in iPSCs derived from patients' fibroblasts.^{130–132} Moretti et al.¹³⁰ were the first to study iPSC-CMs obtained from an LQT1 patient carrying the *KCNQ1-R190Q* mutation, demonstrating that the cells recapitulated the electrophysiological features of the disease (i.e. altered I_{Ks} activation and deactivation resulting in prolonged APD and increased susceptibility to catecholamine-induced tachyarrhythmia). Itzhaki et al.¹³¹ created iPSC-CMs from an LQT2 patient, carrying the *KCNH2-A614V* missense mutation, again recapitulating clinical features and showing the usefulness of the system to evaluate potential pharmacologic treatments. More recently, a drug repurposing strategy was tested in LQT2 iPSC-CMs. Specifically, the drug Lumacaftor, already approved for clinical use in patients with cystic fibrosis harbouring the most common mutation for this disease (CFTR-Phe508 deletion), was tested in LQT2 cardiomyocytes carrying mutations with or without trafficking defect.¹³³ Lumacaftor corrects the trafficking defect induced by the CFTR-Phe508 deletion, through a still unknown mechanism, thus it was hypothesized that it might also correct trafficking defects caused by *KCNH2* mutations ('class 2' mutations).¹³³ Indeed, Lumacaftor reduced the field potential duration (analogous to QT interval at the cellular level) only in iPSC-CMs with trafficking deficient *KCNH2* mutations.¹³³ A Phase II clinical study on two adult patients in whose cardiomyocytes the drug proved efficacious found that the drug shortened QTc by ~30 ms, an encouraging effect but less than expected from data with iPSC-CMs.¹³⁴ Larger clinical studies are clearly warranted to provide more conclusive evidence of the efficacy of Lumacaftor in LQT2.

3.6 Novel therapies

Two novel therapeutic approaches for LQTS have been studied in recent years. Molecules that correct protein trafficking defects represent the first approach. Despite being effective in vitro, their major limitation is their non-specificity and toxicity, and thus, are not suitable for clinical practice.¹³⁵ The only promising drug of this group so far identified is Lumacaftor, that was originally discovered and validated for cystic fibrosis, and later proved to rescue *HERG* trafficking defects in vitro¹³² and to reduce the QTc in the first two LQT2 patients in which it was tested.¹³³ Another strategy tested successfully in iPSC-CMs was the allele-specific RNA interference rescues.¹³⁶ However, applying an RNA-based drug to patients will require more effort. The second approach is represented

by clinically untested compounds that activate I_{Ks} or I_{Kr} . ML277 is a selective *KCNQ1* activator¹³⁷ that enhances I_{Ks} by stabilizing the activated state of the channel¹³⁸ and shortens APD of patient iPSC-CMs-harboring a loss of function mutation in *KCNQ1*.¹³⁹ Several I_{Kr} activators have been identified, such as RPR-260243 that slows the rate of Kv11.1 channel deactivation,¹⁴⁰ and several compounds that markedly enhance outward I_{Kr} by inhibiting channel inactivation, including ICA-105574,¹⁴¹ ML-T531,¹⁴² and AZSMO-23.¹⁴³ PD-118057 increases channel open probability of Kv11.1 and shifts its voltage dependence of inactivation to more positive potentials.¹⁴⁴ NS1643 increases both I_{Kr} peak and tail currents¹⁴⁵ and shortens APD and QT in transgenic LQT2 rabbits.¹⁴⁶ However, all of these compounds affect cardiac ion channels other than Kv11.1 and it is also unclear whether their effects on I_{Kr} can be limited to prevent excessive (and thus potentially pro-arrhythmic) shortening of the QT interval.

4. Short QT syndrome

4.1 Clinical presentation

SQTS is a rare, but potentially lethal congenital channelopathy characterized by accelerated cardiac repolarization and shortened QT interval (QTc < 320–340 ms).^{147,148} The phenotype of SQTS patients is highly variable—ranging from completely asymptomatic to those presenting with various arrhythmia-associated symptoms such as dizziness, syncope, or even SCD.¹⁴⁸ In the worst cases, patients may develop SCD in infancy (sudden infant death syndrome).¹⁴⁹ Forty percent of patients present with SCD due to VT/VF as their initial symptom.¹⁴⁸ Of note, gender differences in the arrhythmia free survival over longer follow-up were reported with significantly higher VT/VF/SCD rates among female SQTS patients than males.^{148,150}

In addition to ventricular arrhythmia, patients are prone to develop early-onset, familial AF,¹⁴⁷ with variable AF prevalence reported in different SQTS families ranging from 10% to over 60%^{151,152} and pronounced genotype differences in AF risk.¹⁵³ The reduced penetrance with a clinically manifest phenotype observed in (only) around 80% of mutation-positive subjects¹⁵³ and the marked variability in disease phenotype,¹⁵⁴ indicate a potential role of genetic modifiers and remodelling as determinants of disease severity.

4.2 Molecular mechanisms

Since its first description in 2000,¹⁵⁵ causative mutations have been identified in six different genes: SQTS types 1–3 are associated with gain-of-function mutations in *KCNH2*, *KCNQ1*, and *KCNJ2*, encoding Kv11.1, Kv7.1, and Kir2.1 channels respectively^{156–158} (Figure 3), whereas SQTS types 4–6 are caused by loss-of-function mutations in Ca^{2+} channels. Mechanisms of K^{+} channel gain of function include reduced inactivation, faster activation, slowed deactivation or constitutive opening (failure to close). In SQT1, missense mutations such as N588K¹⁵⁶ or T681I¹⁵⁹ in Kv11.1 cause a dramatic shift (e.g. +50 mV for T618I) in the voltage required for half-maximal channel inactivation, resulting in a pronounced enhancement of outward I_{Kr} during the plateau phase of the action potential and a markedly shorter APD. In addition to faster ventricular repolarization, SQT2 is characterized by AF and sinus bradycardia and is caused by missense mutations in Kv7.1 such as V307L¹⁵⁷ that enhance the rate of channel activation, or V141M¹⁵⁴ that greatly slows deactivation (channel closure) by altering voltage sensor-pore coupling.¹⁶⁰ SQT3 caused by missense mutations in Kir2.1 (e.g. D172N, V93I, E299V) is characterized by tall and asymmetrical T waves^{39,158,161} and AF in many,

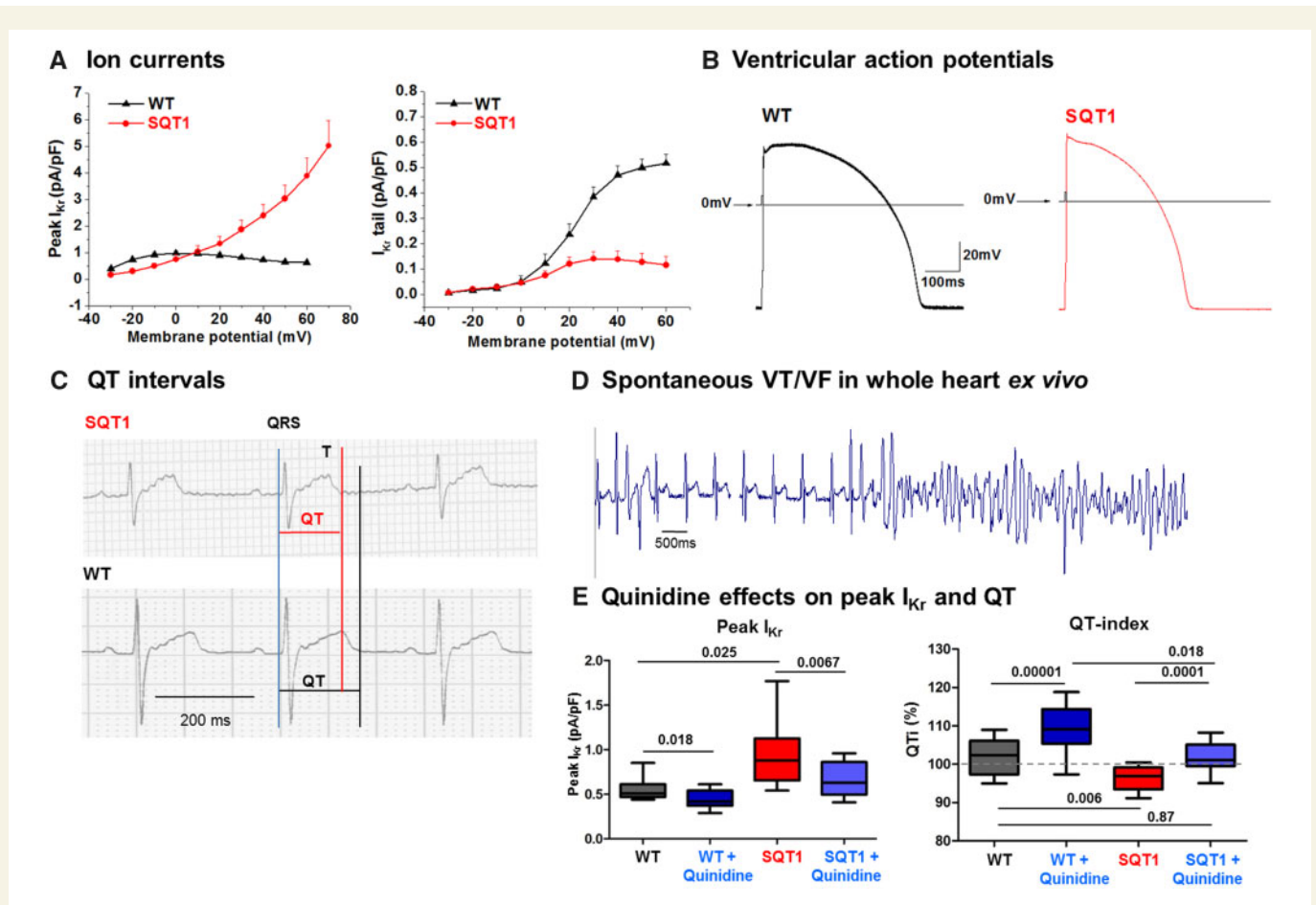


Figure 4 Transgenic SQTS rabbits. (A) Increased peak I_{Kr} , due to impaired inactivation at positive membrane voltages (left panel) and decreased I_{Kr} tail (right panel) in SQT1 cardiomyocytes. (B) Representative cellular action potential recordings in ventricular cardiomyocytes indicating shortened APD in SQT1. (C) Representative surface ECG recordings indicating shortened QT interval in SQT1. (D) Episode of spontaneous VT in Langendorff-perfused SQT1 heart ex vivo. (E) Quinidine reduces peak I_{Kr} in SQT1 cardiomyocytes thereby normalizing the heart rate corrected QT-index (from 95% to 100%) (modified with permission from Odening et al.¹⁶⁹).

but not all patients. These missense mutations all increase outward currents (i.e. reduce extent of inward rectification) conducted by Kir2.1 channels. The SQT3-associated gain of function mutation in Kir2.1, K346T enhances cardiac I_{K1} amplitude by reducing channel protein ubiquitylation and its interaction with caveolin 2, effects that enhance channel targeting to, and stability in, the surface membrane.¹⁶²

4.3 Diagnosis and risk stratification

The diagnostic work-up includes (repetitive) 12-lead ECG, 24-h Holter ECG as well as exercise testing. The 2015 European Society of Cardiology (ESC) Guidelines for the prevention of SCD¹⁶³ suggest that a diagnosis should be reached in all patients with a QTc ≤ 340 ms, while when the QTc is between 340 and 360 ms, SQTS can be diagnosed only in the presence of at least one additional criterion that could be either presence of a pathogenic mutation, family history of SQTS, family history of SCD below age 40 or survival of a VT/VF episode in the absence of heart disease. The Heart Rhythm Society/European Heart Rhythm Association/Asia Pacific Heart Rhythm Society expert consensus statement on the diagnosis and management of patients with inherited primary arrhythmia syndrome⁷⁹ has suggested a slightly lower cut-off (QTc ≤ 330 ms). However, it is worth noting that for QTc to be diagnostic it

should always be below the indicated cut-off value and not only in bradycardic conditions and thus, a 12-lead 24-h Holter evaluation is very important. Another typical feature of affected individuals is the reduced rate-adaptation of QT during exercise testing (more shallow QT/HR relationship) with an absolute QT shortening that should be <60 ms.¹⁶⁴ Finally, other ECG characteristics that could support the diagnosis are the PR depression and a JTp between 90 and 120 ms.¹⁶⁵

Importantly, to date, reliable risk stratification strategies to identify SQTS patients at risk for lethal arrhythmias/SCD are lacking. Inducibility of AF and VT/VF during electrophysiological studies is increased (60%)¹⁶⁶, however, its clinical relevance for risk stratification remains unclear. Scoring systems, which include clinical parameters, ECG criteria and/or electrophysiological work-up (including programmed ventricular stimulation) have not yet helped to identify pro-arrhythmic risk factors and high-risk patients.^{147,148} The only strong predictor of recurrent ventricular arrhythmias is survived/resuscitated cardiac arrest.¹⁴⁸

4.4 Cellular and animal models

Despite the first description of SQTS in 2000,¹⁶⁷ our knowledge on arrhythmia mechanisms, triggers, and consequences for risk stratification is limited, likely due to the small number of SQTS cases (<200)

characterized to date. The lack of knowledge regarding this disorder increases the importance for the development of cellular and animal models that recapitulate the SQTs phenotype to elucidate arrhythmogenic mechanisms, to identify risk factors and to develop novel therapies.

Until recently, only drug-induced SQTs animal models were available with limited translational potential. The first available genetic animal model for SQTs was the zebrafish model *reggae* (expressing the missense mutation L499P in zERG) that results in similar dysfunction in I_{Kr} biophysical properties as that observed in human SQT1¹⁶⁸ as well as a shortened QT. Recently a transgenic rabbit model for SQT1 carrying the disease-specific human mutation *KCNH2*-N588K was generated and characterized.¹⁶⁹ This SQT1 rabbit model mimics the human disease phenotype on all levels (from ion current, cellular, tissue, and whole-heart levels to *in vivo*) with shortened QT, more shallow QT/RR slope, shortened atrial and ventricular refractoriness and APD, as well as increased VT/VF and AF inducibility. This phenotype results from an increase in steady-state I_{Kr} due to impaired channel inactivation¹⁶⁹ (Figure 4). Cellular experiments revealed that the multi-channel blocker quinidine normalized QT and APD in SQT1 rabbits by reducing the pathologically enhanced I_{Kr} . Electrical remodelling was also observed in I_{K1} and I_{Ks} , effects that contribute to the electrical disease phenotype. These unanticipated effects on other currents warrants further investigation and may be causatively linked to phenotypic variability in human subjects.

The first human iPSC-CM genetic models of SQT1 were developed from human skin fibroblasts of different SQT1 patients carrying *KCNH2*-N588K or *KCNH2*-T618I missense mutations.^{170–172} These SQT1 iPSC-CMs resemble the expected patient's cellular phenotype with shortened APD, and spontaneous arrhythmic events. In confluent two-dimensional iPSC-derived cardiac cell sheets of *KCNH2*-N588K iPSC-CMs, arrhythmogenesis could be visualized, revealing increased inducibility of sustained spiral waves, accelerated and stabilized rotors, increased rotor rotation frequency, increased rotor curvature, and increased rotor complexity.¹⁷⁰ Treating the cells with quinidine abolished the arrhythmic events and prolonged APD,^{171,172} indicating that SQT1 iPSC-CMs are useful tools for further mechanistic and therapeutic studies.

4.5 Traditional therapies

As SQTs survivors of SCD continue to be at high risk for developing recurrent ventricular arrhythmias,¹⁴⁸ ICDs are the standard, guideline-recommended therapy for these patients.¹⁷³ However, due to the short QT and the peaked T-wave morphology, the risk for T-wave oversensing-induced inappropriate shocks are relatively high in SQTs patients.¹⁷⁴ Although appropriate shocks are documented in up to 17.5% of SQTs patients, the rate of complications is even higher with up to 54% in patients during a median follow-up time of 67.4 months.¹⁷⁵ Supportive drug therapies may provide additional protective effects, as recently shown for hydroquinidine that prolonged/normalized the QT interval and effectively prevented ventricular arrhythmias in SQTs patients.¹⁷⁶ However, known side effects of hydroquinidine such as gastrointestinal intolerance may lead to non-compliance in many patients, and the drug may not be effective in all genotypes - as previously described for other drugs such as sotalol.^{177,178}

4.6 Novel therapies

SQTs caused by a gain of function mutations in *Kv11.1* or *Kv7.1* channels could conceivably be treated with blockers or gating modulators that reduce net outward conductance of the mutant channel or other K^+

channels that contribute to action potential repolarization. Already approved drugs may have utility in SQTs. Examples include chloroquine for SQT3 because of its ability to block both wild type and mutant *Kir2.1* channels,¹⁷⁹ or a combination of K^+ and Na^+ channel blockers that together prolong both effective refractory period and wavelength for re-entry.¹⁸⁰

5. Conclusions

Normal K^+ channel function is essential to maintain electrical stability in the heart. Gene mutations that alter the assembly, trafficking, turnover or gating of cardiac K^+ channels can cause LQTS, SQTs, J-wave syndromes and AF. Although remarkable progress in our understanding of the genetic and physiological basis of these inheritable K^+ channelopathies have been elucidated in the past 25 years,

mechanism-based therapies are largely missing. We anticipate that a more complete understanding of the molecular basis of mutation-induced channel dysfunction, additional transgenic animal models, as well as advances in gene-therapy techniques will provide insights that yield novel arrhythmia-specific, and in some cases, channel mutation-specific therapies in the future.

Conflict of interest: none declared.

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References

- Schmitt N, Grunnet M, Olesen SP. Cardiac potassium channel subtypes: new roles in repolarization and arrhythmia. *Physiol Rev* 2014;**94**:609–653.
- Barhanin J, Lesage F, Guillemare E, Fink M, Lazdunski M, Romey G. KvLQT1 and Isk (minK) proteins associate to form the I_{Ks} cardiac potassium channel. *Nature* 1996;**384**:78–80.
- Tristani-Firouzi M, Sanguinetti MC. Voltage-dependent inactivation of the human K^+ channel KvLQT1 is eliminated by association with minimal K^+ channel (minK) subunits. *J Physiol* 1998;**510**:37–45.
- Sanguinetti MC, Jiang C, Curran ME, Keating MT. A mechanistic link between an inherited and an acquired cardiac arrhythmia: *HERG* encodes the I_{Kr} potassium channel. *Cell* 1995;**81**:299–307.
- Trudeau M, Warmke JW, Ganetzky B, Robertson GA. *HERG*, A human inward rectifier in the voltage-gated potassium channel family. *Science* 1995;**269**:92–95.
- Jones EM, Roti Roti EC, Wang J, Delfosse SA, Robertson GA. Cardiac I_{Kr} channels minimally comprise *HERG* 1a and 1b subunits. *J Biol Chem* 2004;**279**:44690–44694.
- Feng J, Xu D, Wang Z, Nattel S. Ultrarapid delayed rectifier current inactivation in human atrial myocytes: properties and consequences. *Am J Physiol* 1998;**275**:H1717–H1725.
- Ravens U, Wettwer E. Ultra-rapid delayed rectifier channels: molecular basis and therapeutic implications. *Cardiovasc Res* 2011;**89**:776–785.
- Brahmajothi MV, Campbell DL, Rasmusson RL, Morales MJ, Trimmer JS, Nerbonne JM, Strauss HC. Distinct transient outward potassium current (I_{to}) phenotypes and distribution of fast-inactivating potassium channel alpha subunits in ferret left ventricular myocytes. *J Gen Physiol* 1999;**113**:581–600.
- Rosati B, Pan Z, Lyden S, Wang HS, Cohen I, Dixon JE, McKinnon D. Regulation of KChIP2 potassium channel beta subunit gene expression underlies the gradient of transient outward current in canine and human ventricle. *J Physiol* 2001;**533**:119–125.
- Kim LA, Furst J, Butler MH, Xu S, Grigorieff N, Goldstein SA. Composition of I_{to} channels: Kv4.2-KChIP2 complexes carry four subunits of each type. *J Biol Chem* 2004;**279**:5549–5554.
- Nichols CG, Lee SJ. Polyamines and potassium channels: a 25-year romance. *J Biol Chem* 2018;**293**:18779–18788.
- Rohacs T, Chen J, Prestwich GD, Logothetis DE. Distinct specificities of inwardly rectifying K^+ channels for phosphoinositides. *J Biol Chem* 1999;**274**:36065–36072.
- Anumonwo JM, Lopatin AN. Cardiac strong inward rectifier potassium channels. *J Mol Cell Cardiol* 2010;**48**:45–54.

15. Krapivinsky G, Gordon EA, Wickman K, Velimirovic B, Krapivinsky L, Clapham DE. The G-protein-gated atrial K^+ channel $I_{K_{ACH}}$ is a heteromultimer of two inwardly rectifying K^+ -channel proteins. *Nature* 1995;**374**:135–141.
16. Nichols CG. Adenosine triphosphate-sensitive potassium currents in heart disease and cardioprotection. *Card Electrophysiol Clin* 2016;**8**:323–335.
17. Zhang H, Flagg TP, Nichols CG. Cardiac sarcolemmal K_{ATP} channels: latest twists in a questing tale! *J Mol Cell Cardiol* 2010;**48**:71–75.
18. Zhang XD, Lieu DK, Chiamvimonvat N. Small-conductance Ca^{2+} -activated K^+ channels and cardiac arrhythmias. *Heart Rhythm* 2015;**12**:1845–1851.
19. Lee CH, MacKinnon R. Activation mechanism of a human SK-calmodulin channel complex elucidated by cryo-EM structures. *Science* 2018;**360**:508–513.
20. Skibsbjerg L, Poulet C, Diness JG, Bentzen BH, Yuan L, Kappert U, Matschke K, Wettwer E, Ravens U, Grunnet M, Christ T, Jespersen T. Small-conductance calcium-activated potassium (SK) channels contribute to action potential repolarization in human atria. *Cardiovasc Res* 2014;**103**:156–167.
21. Smith CO, Wang YT, Nadtchuy SM, Miller JH, Jonas EA, Dirksen RT, Nehrke K, Brookes PS. Cardiac metabolic effects of $K_{Na}1.2$ channel deletion and evidence for its mitochondrial localization. *FASEB J* 2018;**32**:6135–6149.
22. Smith CO, Nehrke K, Brookes PS. The Slo(w) path to identifying the mitochondrial channels responsible for ischemic protection. *Biochem J* 2017;**474**:2067–2094.
23. Gaborit N, Le Bouter S, Szuts V, Varro A, Escande D, Nattel S, Demolombe S. Regional and tissue specific transcript signatures of ion channel genes in the non-diseased human heart. *J Physiol* 2007;**582**:675–693.
24. Rinne S, Kiper AK, Schlichthorl G, Dittmann S, Netter MF, Limberg SH, Silbernagel N, Zuzarte M, Moosdorf R, Wulf H, Schulze-Bahr E, Rolfes C, Decher N. TASK-1 and TASK-3 may form heterodimers in human atrial cardiomyocytes. *J Mol Cell Cardiol* 2015;**81**:71–80.
25. Chai S, Wan X, Nassal DM, Liu H, Moravec CS, Ramirez-Navarro A, Deschênes I. Contribution of two-pore K^+ channels to cardiac ventricular action potential revealed using human iPSC-derived cardiomyocytes. *Am J Physiol Heart Circ Physiol* 2017;**312**:H1144–H1153.
26. Schmidt C, Wiedmann F, Kallenberger SM, Ratte A, Schulte JS, Scholz B, Müller FU, Voigt N, Zafeiriou MP, Ehrlich JR, Tochtermann U, Veres G, Ruhparwar A, Karck M, Katus HA, Thomas D. Stretch-activated two-pore-domain (K_2P) potassium channels in the heart: focus on atrial fibrillation and heart failure. *Prog Biophys Mol Biol* 2017;**130**:233–243.
27. Marx SO, Kurokawa J, Reiken S, Motoike H, D'Armiento J, Marks AR, Kass RS. Requirement of a macromolecular signaling complex for beta adrenergic receptor modulation of the KCNQ1-KCNE1 potassium channel. *Science* 2002;**295**:496–499.
28. Terrenoire C, Houslay MD, Baillie GS, Kass RS. The cardiac I_{Ks} potassium channel macromolecular complex includes the phosphodiesterase PDE4D3. *J Biol Chem* 2009;**284**:9140–9146.
29. Li Y, Chen L, Kass RS, Dessauer CW. The A-kinase anchoring protein Yotiao facilitates complex formation between adenylyl cyclase type 9 and I_{Ks} potassium channel in heart. *J Biol Chem* 2012;**287**:29815–29824.
30. Loussouarn G, Park KH, Bellocq C, Baro I, Charpentier F, Escande D. Phosphatidylinositol-4,5-bisphosphate, PIP_2 , controls KCNQ1/KCNE1 voltage-gated potassium channels: a functional homology between voltage-gated and inward rectifier K^+ channels. *EMBO J* 2003;**22**:5412–5421.
31. Cui J. Voltage-dependent gating: novel insights from KCNQ1 channels. *Biophys J* 2016;**110**:14–25.
32. Sun J, MacKinnon R. Cryo-EM structure of a KCNQ1/CaM complex reveals insights into congenital long QT syndrome. *Cell* 2017;**169**:1042–1050.
33. Chen L, Sampson KJ, Kass RS. Cardiac delayed rectifier potassium channels in health and disease. *Card Electrophysiol Clin* 2016;**8**:307–322.
34. Grandi E, Sanguinetti MC, Bartos DC, Bers DM, Chen-lzu Y, Chiamvimonvat N, Colecraft HM, Delisle BP, Heijman J, Navedo MF, Noskov S, Proenza C, Vandenberg JJ, Yarov-Yarovoy V. Potassium channels in the heart: structure, function and regulation. *J Physiol* 2017;**595**:2209–2228.
35. Oyler J, Maljevic S, Scheffer IE, Berkovic SF, Petrou S, Reid CA. Ion channels in genetic epilepsy: from genes and mechanisms to disease-targeted therapies. *Pharmacol Rev* 2018;**70**:142–173.
36. Rorsman P, Ashcroft FM. Pancreatic beta-cell electrical activity and insulin secretion: of mice and men. *Physiol Rev* 2018;**98**:117–214.
37. D'Adamo MC, Hasan S, Guglielmi L, Servetini I, Cenciari M, Catacuzzeno L, Franciolini F. New insights into the pathogenesis and therapeutics of episodic ataxia type 1. *Front Cell Neurosci* 2015;**9**:317.
38. Levin MD, Zhang H, Uchida K, Grange DK, Singh GK, Nichols CG. Electrophysiologic consequences of K_{ATP} gain of function in the heart: conduction abnormalities in Cantu syndrome. *Heart Rhythm* 2015;**12**:2316–2324.
39. Xia M, Jin Q, Bendahhou S, He Y, Larroque M-M, Chen Y, Zhou Q, Yang Y, Liu Y, Liu B, Zhu Q, Zhou Y, Lin J, Liang B, Li L, Dong X, Pan Z, Wang R, Wan H, Qiu W, Xu W, Eurlings P, Barhanin J, Chen Y. A Kir2.1 gain-of-function mutation underlies familial atrial fibrillation. *Biochem Biophys Res Commun* 2005;**332**:1012–1019.
40. Olson TM, Alekseev AE, Liu XK, Park S, Zingman LV, Bienengraeber M, Sattiraju S, Ballew JD, Jahangir A, Terzic A. Kv1.5 channelopathy due to KCNA5 loss-of-function mutation causes human atrial fibrillation. *Hum Mol Genet* 2006;**15**:2185–2191.
41. Drabkin M, Zilberberg N, Menahem S, Mulla W, Halperin D, Yogev Y, Wormser O, Perez Y, Kadir R, Etzion Y, Katz A, Birk OS. Nocturnal atrial fibrillation caused by mutation in KCND2, encoding pore-forming (alpha) subunit of the cardiac Kv4.2 potassium channel. *Circ Genom Precis Med* 2018;**11**:e002293.
42. Olesen MS, Refsgaard L, Holst AG, Larsen AP, Grubb S, Haunso S, Svendsen JH, Olesen SP, Schmitt N, Calloe K. A novel KCND3 gain-of-function mutation associated with early-onset of persistent lone atrial fibrillation. *Cardiovasc Res* 2013;**98**:488–495.
43. Jabbari J, Olesen MS, Holst AG, Nielsen JB, Haunso S, Svendsen JH. Common polymorphisms in KCNJ5 are associated with early-onset lone atrial fibrillation in Caucasians. *Cardiology* 2011;**118**:116–120.
44. Yamada N, Asano Y, Fujita M, Yamazaki S, Inanobe A, Matsuura N, Kobayashi H, Ohno S, Ebana Y, Tsukamoto O, Ishino S, Takuwa A, Kioka H, Yamashita T, Hashimoto N, Zankov DP, Shimizu A, Asakura M, Asanuma H, Kato H, Nishida Y, Miyashita Y, Shinomiya H, Naiki N, Hayashi K, Makiyama T, Ogita H, Miura K, Ueshima H, Komuro I, Yamagishi M, Horie M, Kawakami K, Furukawa T, Koizumi A, Kurachi Y, Sakata Y, Minamino T, Kitakaze M, Takashima S. Mutant KCNJ3 and KCNJ5 potassium channels as novel molecular targets in bradyarrhythmias and atrial fibrillation. *Circulation* 2019;**139**:2157–2169.
45. Dobrev D, Friedrich A, Voigt N, Jost N, Wettwer E, Christ T, Knaut M, Ravens U. The G protein-gated potassium current $I_{K_{ACH}}$ is constitutively active in patients with chronic atrial fibrillation. *Circulation* 2005;**112**:3697–3706.
46. Kuß J, Stallmeyer B, Goldstein M, Rinné S, Pees C, Zumhagen S, Seeböhm G, Decher N, Pott L, Kienitz M-C, Schulze-Bahr E. Familial sinus node disease caused by a gain of GIRK (G-protein activated inwardly rectifying $K(+) channel$) channel function. *Circ Genom Precis Med* 2019;**12**:e002238.
47. Lundby A, Ravn LS, Svendsen JH, Hauns S, Olesen SP, Schmitt N. KCNE3 mutation V17M identified in a patient with lone atrial fibrillation. *Cell Physiol Biochem* 2008;**21**:047–054.
48. Ravn LS, Aizawa Y, Pollevick GD, Hofman-Bang J, Cordeiro JM, Dixon U, Jensen G, Wu Y, Burashnikov E, Haunso S, Guericoff A, Hu D, Svendsen JH, Christiansen M, Antzelevitch C. Gain of function in I_{Ks} secondary to a mutation in KCNE5 associated with atrial fibrillation. *Heart Rhythm* 2008;**5**:427–435.
49. Abbott GW. KCNE4 and KCNE5: K^+ channel regulation and cardiac arrhythmogenesis. *Gene* 2016;**593**:249–260.
50. Liang B, Soka M, Christensen AH, Olesen MS, Larsen AP, Knop FK, Wang F, Nielsen JB, Andersen MN, Humphreys D, Mann SA, Huttner IG, Vandenberg JJ, Svendsen JH, Haunso S, Preiss T, Seeböhm G, Olesen SP, Schmitt N, Fatkin D. Genetic variation in the two-pore domain potassium channel, TASK-1, may contribute to an atrial substrate for arrhythmogenesis. *J Mol Cell Cardiol* 2014;**67**:69–76.
51. Antzelevitch C, Yan GX, Ackerman MJ, Borggreffe M, Corrado D, Guo J, Gussak I, Hasdemir C, Horie M, Huikuri H, Ma C, Morita H, Nam GB, Sacher F, Shimizu W, Viskin S, Wilde AA. J-Wave syndromes expert consensus conference report: emerging concepts and gaps in knowledge. *J Arrhythm* 2016;**32**:315–339.
52. Giudicessi JR, Ye D, Tester DJ, Crotti L, Mugione A, Nesterenko VV, Albertson RM, Antzelevitch C, Schwartz PJ, Ackerman MJ. Transient outward current (I_{to}) gain-of-function mutations in the KCND3-encoded Kv4.3 potassium channel and Brugada syndrome. *Heart Rhythm* 2011;**8**:1024–1032.
53. Haissaguerre M, Chatel S, Sacher F, Weerasooriya R, Probst V, Loussouarn G, Horlitz M, Liersch R, Schulze-Bahr E, Wilde A, Kaab S, Koster J, Rudy Y, Le Marec H, Schott JJ. Ventricular fibrillation with prominent early repolarization associated with a rare variant of KCNJ8/KATP channel. *J Cardiovasc Electrophysiol* 2009;**20**:93–98.
54. Delpont E, Cordeiro JM, Nunez L, Thomsen PE, Guericoff A, Pollevick GD, Wu Y, Kanters JK, Larsen CT, Hofman-Bang J, Burashnikov E, Christiansen M, Antzelevitch C. Functional effects of KCNE3 mutation and its role in the development of Brugada syndrome. *Circ Arrhythm Electrophysiol* 2008;**1**:209–218.
55. Takayama K, Ohno S, Ding WG, Ashihara T, Fukumoto D, Wada Y, Makiyama T, Kise H, Hoshiai M, Matsuura H, Horie M. A de novo gain-of-function KCND3 mutation in early repolarization syndrome. *Heart Rhythm* 2019;**16**:1698–1706.
56. Schwartz P, Crotti L. Long QT and short QT syndromes. In D Zipes, J Jalife, W (eds). *Stevenson Cardiac Electrophysiology: From Cell to Bedside*. Philadelphia, PA: Elsevier/Saunders, 2017. pp. 893–904.
57. Ward OC. A new familial cardiac syndrome in children. *J Ir Med Assoc* 1964;**54**:103–106.
58. Schwartz PJ, Stramba-Badiale M, Crotti L, Pedrazzini M, Besana A, Bosi G, Gabbiani F, Goulene K, Insolia R, Mannarino S, Mosca F, Napolitano L, Rimini A, Rosati E, Salice P, Spazzolini C. Prevalence of the congenital long-QT syndrome. *Circulation* 2009;**120**:1761–1767.
59. Schwartz PJ, Spazzolini C, Crotti L, Bathen J, Amlie JP, Timothy K, Shkolnikova M, Berul CI, Bitner-Glindzic M, Toivonen L, Horie M, Schulze-Bahr E, Denjoy I. The Jervell and Lange-Nielsen syndrome: natural history, molecular basis, and clinical outcome. *Circulation* 2006;**113**:783–790.
60. Sanguinetti MC, Curran ME, Zou A, Shen J, Specter PS, Atkinson DL, Keating MT. Coassembly of K_vLQT1 and $minK$ (IsK) proteins to form cardiac I_{Ks} potassium channel. *Nature* 1996;**384**:80–83.
61. Abbott GW, Sesti F, Splawski I, Buck ME, Lehmann MH, Timothy KW, Keating MT, Goldstein S. MiRP1 forms I_{Kr} potassium channels with HERG and is associated with cardiac arrhythmia. *Cell* 1999;**97**:175–187.

62. Roberts JD, Asaki SY, Mazzanti A, Bos JM, Tuleta I, Muir AR, Crotti L, Krahn AD, Kutlyifa V, Shoemaker MB, Johnsrude CL, Aiba T, Marcondes L, Baban A, Udupa S, Dechert B, Fischbach P, Knight LM, Vittinghoff E, Kukavica D, Stallmeyer B, Giudicessi JR, Spazzolini C, Shimamoto K, Tadmor R, Cadrin-Tourigny J, Duff HJ, Simpson CS, Roston TM, Wijeyeratne YD, El Hajjaji I, Yousif MD, Gula LJ, Leong-Sit P, Chavali N, Landstrom AP, Marcus GM, Dittmann S, Wilde A, Behr ER, Tfelt-Hansen J, Scheinman MM, Perez MV, Kaski JP, Gow RM, Drago F, Aziz PF, Abrams DJ, Gollob MH, Skinner JR, Kaufman ES, Roden DM, Zareba W, Schwartz PJ, Schulze-Bahr E, Etheridge SP, Priori SG, Ackerman MJ. An international multicenter evaluation of type 5 long QT syndrome: a low penetrant primary arrhythmic condition. *Circulation* 2020;**141**:429–439.
63. Bohnen MS, Peng G, Robey SH, Terrenoire C, Iyer V, Sampson KJ, Kass RS. Molecular pathophysiology of congenital long QT syndrome. *Physiol Rev* 2017;**97**: 89–134.
64. Dvir M, Strulovich R, Sachyani D, Ben-Tal Cohen I, Haitin Y, Dessauer C, Pongs O, Kass R, Hirsch JA, Attali B. Long QT mutations at the interface between KCNQ1 helix C and KCNE1 disrupt I_{Ks} regulation by PKA and PIP₂. *J Cell Sci* 2014;**127**: 3943–3955.
65. Schwartz PJ, Priori SG, Spazzolini C, Moss AJ, Vincent GM, Napolitano C, Denjoy I, Guicheney P, Breithardt G, Keating MT, Towbin JA, Beggs AH, Brink P, Wilde AA, Toivonen L, Zareba W, Robinson JL, Timothy KW, Corfield V, Wattanasirichaigoon D, Corbett C, Haverkamp W, Schulze-Bahr E, Lehmann MH, Schwartz K, Coumel P, Bloise R. Genotype-phenotype correlation in the long-QT syndrome: gene-specific triggers for life-threatening arrhythmias. *Circulation* 2001;**103**:89–95.
66. Buber J, Mathew J, Moss AJ, Hall WJ, Barsheshet A, McNitt S, Robinson JL, Zareba W, Ackerman MJ, Kaufman ES, Luria D, Eldar M, Towbin JA, Vincent M, Goldenberg I. Risk of recurrent cardiac events after onset of menopause in women with congenital long-QT syndrome types 1 and 2. *Circulation* 2011;**123**:2784–2791.
67. Schwartz PJ, Crotti L. QTc behavior during exercise and genetic testing for the long-QT syndrome. *Circulation* 2011;**124**:2181–2184.
68. Nador F, Beria G, De Ferrari GM, Stramba-Badiale M, Locati EH, Lotto A, Schwartz PJ. Unsuspected echocardiographic abnormality in the long QT syndrome. Diagnostic, prognostic, and pathogenetic implications. *Circulation* 1991;**84**: 1530–1542.
69. Haugaa KH, Amlie JP, Berge KE, Lerer TP, Smiseth OA, Edvardsen T. Transmural differences in myocardial contraction in long-QT syndrome: mechanical consequences of ion channel dysfunction. *Circulation* 2010;**122**:1355–1363.
70. Brado J, Dechant MJ, Menza M, Komancsek A, Lang CN, Bugger H, Foell D, Jung BA, Stiller B, Bode C, Odening KE. Phase-contrast magnet resonance imaging reveals regional, transmural, and base-to-apex dispersion of mechanical dysfunction in patients with long QT syndrome. *Heart Rhythm* 2017;**14**:1388–1397.
71. ter Bekke RM, Haugaa KH, van den Wijngaard A, Bos JM, Ackerman MJ, Edvardsen T, Volders PG. Electromechanical window negativity in genotyped long-QT syndrome patients: relation to arrhythmia risk. *Eur Heart J* 2015;**36**:179–186.
72. Jervell A, Lange-Nielsen F. Congenital deaf-mutism, functional heart disease with prolongation of the Q-T interval and sudden death. *Am Heart J* 1957;**54**:59–68.
73. Nguyen HL, Pieper GH, Wilders R. Andersen-Tawil syndrome: clinical and molecular aspects. *Int J Cardiol* 2013;**170**:1–16.
74. Delannoy E, Sacher F, Maury P, Mabo P, Mansourati J, Magnin I, Camous JP, Tournant G, Rendu E, Kyndt F, Haissaguerre M, Bezieau S, Guyomarch B, Le Marec H, Fressart V, Denjoy I, Probst V. Cardiac characteristics and long-term outcome in Andersen-Tawil syndrome patients related to KCNJ2 mutation. *Europace* 2013;**15**: 1805–1811.
75. Tristani-Firouzi M, Jensen JL, Donaldson MR, Sansone V, Meola G, Hahn A, Bendahhou S, Kwiecinski H, Fidzianska A, Plaster N, Fu YH, Ptacek LJ, Tawil R. Functional and clinical characterization of KCNJ2 mutations associated with LQT7 (Andersen syndrome). *J Clin Invest* 2002;**110**:381–388.
76. Johnson JN, Hofman N, Haglund CM, Cascino GD, Wilde AA, Ackerman MJ. Identification of a possible pathogenic link between congenital long QT syndrome and epilepsy. *Neurology* 2009;**72**:224–231.
77. Partemi S, Cestele S, Pezzella M, Campuzano O, Paravidino R, Pascali VL, Zara F, Tassinari CA, Striano S, Oliva A, Brugada R, Mantegazza M, Striano P. Loss-of-function KCNH2 mutation in a family with long QT syndrome, epilepsy, and sudden death. *Epilepsia* 2013;**54**:e112–e116.
78. Tiron C, Campuzano O, Perez-Serra A, Mademont I, Coll M, Allegue C, Iglesias A, Partemi S, Striano P, Oliva A, Brugada R. Further evidence of the association between LQT syndrome and epilepsy in a family with KCNQ1 pathogenic variant. *Seizure* 2015;**25**:65–67.
79. Priori SG, Wilde AA, Horie M, Cho Y, Behr ER, Berul C, Blom N, Brugada J, Chiang CE, Huikuri H, Kannankeril P, Krahn A, Leenhardt A, Moss A, Schwartz PJ, Shimizu W, Tomaselli G, Tracy C. HRS/EHRA/APHRS expert consensus statement on the diagnosis and management of patients with inherited primary arrhythmia syndromes: document endorsed by HRS, EHRA, and APHRS in May 2013 and by ACCF, AHA, PACES, and AEP in June 2013. *Heart Rhythm* 2013;**10**:1932–1963.
80. Itoh H, Crotti L, Aiba T, Spazzolini C, Denjoy I, Fressart V, Hayashi K, Nakajima T, Ohno S, Makiyama T, Wu J, Hasegawa K, Mastantuono E, Dagradi F, Pedrazzini M, Yamagishi M, Berthet M, Murakami Y, Shimizu W, Guicheney P, Schwartz PJ, Horie M. The genetics underlying acquired long QT syndrome: impact for genetic screening. *Eur Heart J* 2016;**37**:1456–1464.
81. Mazzanti A, Maragna R, Vacanti G, Monteforte N, Bloise R, Marino M, Braghieri L, Gambelli P, Memmi M, Pagan E, Morini M, Malovini A, Ortiz M, Sacilotto L, Bellazzi R, Monserrat L, Napolitano C, Bagnardi V, Priori SG. Interplay between genetic substrate, QTc duration, and arrhythmia risk in patients with long QT syndrome. *J Am Coll Cardiol* 2018;**71**:1663–1671.
82. Moss AJ, Schwartz PJ, Crampton RS, Locati E, Carleen E. The long QT syndrome: a prospective international study. *Circulation* 1985;**71**:17–21.
83. Spazzolini C, Mullally J, Moss AJ, Schwartz PJ, McNitt S, Ouellet G, Fugate T, Goldenberg I, Jons C, Zareba W, Robinson JL, Ackerman MJ, Benhorin J, Crotti L, Kaufman ES, Locati EH, Qi M, Napolitano C, Priori SG, Towbin JA, Vincent GM. Clinical implications for patients with long QT syndrome who experience a cardiac event during infancy. *J Am Coll Cardiol* 2009;**54**:832–837.
84. Priori SG, Schwartz PJ, Napolitano C, Bloise R, Ronchetti E, Grillo M, Vicentini A, Spazzolini C, Nastoli J, Bottelli G, Folli R, Cappelletti D. Risk stratification in the long-QT syndrome. *N Engl J Med* 2003;**348**:1866–1874.
85. Schwartz PJ, Vanoli E, Crotti L, Spazzolini C, Ferrandi C, Goosen A, Hedley P, Heradien M, Bacchini S, Turco A, La Rovere MT, Bartoli A, George AL Jr, Brink PA. Neural control of heart rate is an arrhythmia risk modifier in long QT syndrome. *J Am Coll Cardiol* 2008;**51**:920–929.
86. Crotti L, Spazzolini C, Porretta AP, Dagradi F, Taravelli E, Petracci B, Vicentini A, Pedrazzini M, La Rovere MT, Vanoli E, Goosen A, Heradien M, George AL Jr, Brink PA, Schwartz PJ. Vagal reflexes following an exercise stress test: a simple clinical tool for gene-specific risk stratification in the long QT syndrome. *J Am Coll Cardiol* 2012;**60**:2515–2524.
87. Westenskow P, Splawski I, Timothy KW, Keating MT, Sanguinetti MC. Compound mutations: a common cause of severe long-QT syndrome. *Circulation* 2004;**109**: 1834–1841.
88. Moss AJ, Zareba W, Kaufman ES, Gattman E, Peterson DR, Benhorin J, Towbin JA, Keating MT, Priori SG, Schwartz PJ, Vincent GM, Robinson JL, Andrews ML, Feng C, Hall WJ, Medina A, Zhang L, Wang Z. Increased risk of arrhythmic events in long-QT syndrome with mutations in the pore region of the human ether-a-go-go-related gene potassium channel. *Circulation* 2002;**105**:794–799.
89. Moss AJ, Shimizu W, Wilde AA, Towbin JA, Zareba W, Robinson JL, Qi M, Vincent GM, Ackerman MJ, Kaufman ES, Hofman N, Seth R, Kamakura S, Miyamoto Y, Goldenberg I, Andrews ML, McNitt S. Clinical aspects of type-1 long-QT syndrome by location, coding type, and biophysical function of mutations involving the KCNQ1 gene. *Circulation* 2007;**115**:2481–2489.
90. Crotti L, Spazzolini C, Schwartz PJ, Shimizu W, Denjoy I, Schulze-Bahr E, Zaklyazminskaya EV, Swan H, Ackerman MJ, Moss AJ, Wilde AA, Horie M, Brink PA, Insolia R, De Ferrari GM, Crimi G. The common long-QT syndrome mutation KCNQ1/A341V causes unusually severe clinical manifestations in patients with different ethnic backgrounds: toward a mutation-specific risk stratification. *Circulation* 2007;**116**:2366–2375.
91. Schwartz PJ, Crotti L, George AL Jr. Modifier genes for sudden cardiac death. *Eur Heart J* 2018;**39**:3925–3931.
92. Crotti L, Lundquist AL, Insolia R, Pedrazzini M, Ferrandi C, De Ferrari GM, Vicentini A, Yang P, Roden DM, George AL Jr, Schwartz PJ. KCNH2-K897T is a genetic modifier of latent congenital long-QT syndrome. *Circulation* 2005;**112**:1251–1258.
93. Amin AS, Giudicessi JR, Tijssen AJ, Spanjaart AM, Reckman YJ, Klemens CA, Tanck MW, Kapplinger JD, Hofman N, Sinner MF, Muller M, Wijnen WJ, Tan HL, Bezzina CR, Creemers EE, Wilde AA, Ackerman MJ, Pinto YM. Variants in the 3' untranslated region of the KCNQ1-encoded Kv7.1 potassium channel modify disease severity in patients with type 1 long QT syndrome in an allele-specific manner. *Eur Heart J* 2012;**33**:714–723.
94. Duchatelet S, Crotti L, Peat RA, Denjoy I, Itoh H, Berthet M, Ohno S, Fressart V, Monti MC, Crocamo C, Pedrazzini M, Dagradi F, Vicentini A, Klug D, Brink PA, Goosen A, Swan H, Toivonen L, Lahtinen AM, Kontula K, Shimizu W, Horie M, George AL Jr, Tregouet DA, Guicheney P, Schwartz PJ. Identification of a KCNQ1 polymorphism acting as a protective modifier against arrhythmic risk in long-QT syndrome. *Circ Cardiovasc Genet* 2013;**6**:354–361.
95. de Villiers CP, van der Merwe L, Crotti L, Goosen A, George AL Jr, Schwartz PJ, Brink PA, Moolman-Smook JC, Corfield VA. AKAP9 is a genetic modifier of congenital long-QT syndrome type 1. *Circ Cardiovasc Genet* 2014;**7**:599–606.
96. Crotti L, Monti MC, Insolia R, Peljo A, Goosen A, Brink PA, Greenberg DA, Schwartz PJ, George AL Jr. NOS1AP is a genetic modifier of the long-QT syndrome. *Circulation* 2009;**120**:1657–1663.
97. Tomas M, Napolitano C, De Giuli L, Bloise R, Subirana I, Malovini A, Bellazzi R, Arking DE, Marban E, Chakravarti A, Spooner PM, Priori SG. Polymorphisms in the NOS1AP gene modulate QT interval duration and risk of arrhythmias in the long QT syndrome. *J Am Coll Cardiol* 2010;**55**:2745–2752.
98. Kolder I, Tanck MW, Postema PG, Barc J, Sinner MF, Zumhagen S, Husemann A, Stallmeyer B, Koopmann TT, Hofman N, Pfeufer A, Lichtner P, Meitinger T, Beckmann BM, Myerburg RJ, Bishopp NH, Roden DM, Kaab S, Wilde AAM, Schott JJ, Schulze-Bahr E, Bezzina CR. Analysis for genetic modifiers of disease severity in patients with long-QT syndrome type 2. *Circ Cardiovasc Genet* 2015;**8**:447–456.
99. Ronchi C, Bernardi J, Mura M, Stefanello M, Badone B, Rocchetti M, Crotti L, Brink P, Schwartz PJ, Gnechchi M, Zaza A. NOS1AP polymorphisms reduce NOS1 activity and interact with prolonged repolarization in arrhythmogenesis. *Cardiovasc Res* 2020;doi:10.1093/cvr/cvaa036.

100. Chockalingam P, Crotti L, Girardengo G, Johnson JN, Harris KM, van der Heijden JF, Hauer RN, Beckmann BM, Spazzolini C, Rordorf R, Rydberg A, Clur SA, Fischer M, van den Heuvel F, Kaab S, Blom NA, Ackerman MJ, Schwartz PJ, Wilde AA. Not all beta-blockers are equal in the management of long QT syndrome types 1 and 2: higher recurrence of events under metoprolol. *J Am Coll Cardiol* 2012;**60**:2092–2099.
101. Bos JM, Crotti L, Rohatgi RK, Castelletti S, Dagradi F, Schwartz PJ, Ackerman MJ. Mexiletine shortens the QT interval in patients with potassium channel-mediated type 2 long QT syndrome. *Circ Arrhythm Electrophysiol* 2019;**12**:e007280.
102. Schwartz PJ. Cardiac sympathetic denervation to prevent life-threatening arrhythmias. *Nat Rev Cardiol* 2014;**11**:346–353.
103. Schwartz PJ, Priori SG, Cerrone M, Spazzolini C, Otero A, Napolitano C, Bloise R, De Ferrari GM, Klersy C, Moss AJ, Zareba W, Robinson JL, Hall WJ, Brink PA, Toivonen L, Epstein AE, Li C, Hu D. Left cardiac sympathetic denervation in the management of high-risk patients affected by the long-QT syndrome. *Circulation* 2004;**109**:1826–1833.
104. Yagishita D, Chui RW, Yamakawa K, Rajendran PS, Ajjola OA, Nakamura K, So EL, Mahajan A, Shivkumar K, Vaseghi M. Sympathetic nerve stimulation, not circulating norepinephrine, modulates T-peak to T-end interval by increasing global dispersion of repolarization. *Circ Arrhythm Electrophysiol* 2015;**8**:174–185.
105. Schwartz PJ, Spazzolini C, Priori SG, Crotti L, Vicentini A, Landolina M, Gasparini M, Wilde AA, Knops RE, Denjoy I, Toivonen L, Monnig G, Al-Fayyadh M, Jordaens L, Borggrefe M, Holmgren C, Brugada P, De Roy L, Hohnloser SH, Brink PA. Who are the long-QT syndrome patients who receive an implantable cardioverter-defibrillator and what happens to them?: data from the European Long-QT Syndrome Implantable Cardioverter-Defibrillator (LQTS ICD) Registry. *Circulation* 2010;**122**:1272–1282.
106. Nerbonne JM. Molecular basis of functional voltage-gated K⁺ channel diversity in the mammalian myocardium. *J Physiol* 2000;**525**:285–298.
107. Salama G, London B. Mouse models of long QT syndrome. *J Physiol* 2007;**578**:43–53.
108. Salata JJ, Jurkiewicz NK, Jow B, Folander K, Guinasso PJ Jr, Raynor B, Swanson R, Fermini B. I_K of rabbit ventricle is composed of two currents: evidence for I_{Ks}. *Am J Physiol* 1996;**271**:H2477–H2489.
109. Brunner M, Peng X, Liu GX, Ren XQ, Ziv O, Choi BR, Mathur R, Hajjiri M, Odening KE, Steinberg E, Folco EJ, Pringa E, Centracchio J, Macharzina RR, Donahay T, Schofield L, Rana N, Kirk M, Mitchell GF, Poppas A, Zehender M, Koren G. Mechanisms of cardiac arrhythmias and sudden death in transgenic rabbits with long QT syndrome. *J Clin Invest* 2008;**118**:2246–2259.
110. Major P, Baczkó I, Hiripi L, Odening KE, Juhász V, Kohajda Z, Horváth A, Seprényi G, Kovács M, Virág L, Jost N, Prorok J, Ördög B, Doleschall Z, Nattel S, Varró A, Bösze Z. A novel transgenic rabbit model with reduced repolarization reserve: long QT syndrome caused by a dominant-negative mutation of the KCNE1 gene. *Br J Pharmacol* 2016;**173**:2046–2061.
111. Odening KE, Kirk M, Brunner M, Ziv O, Lorvidhaya P, Liu GX, Schofield L, Chaves L, Peng X, Zehender M, Choi BR, Koren G. Electrophysiological studies of transgenic long QT type 1 and type 2 rabbits reveal genotype-specific differences in ventricular refractoriness and His conduction. *Am J Physiol Heart Circ Physiol* 2010;**299**:H643–655.
112. Odening KE, Choi BR, Liu GX, Hartmann K, Ziv O, Chaves L, Schofield L, Centracchio J, Zehender M, Peng X, Brunner M, Koren G. Estradiol promotes sudden cardiac death in transgenic long QT type 2 rabbits while progesterone is protective. *Heart Rhythm* 2012;**9**:823–832.
113. Odening KE, Jung BA, Lang CN, Cabrera Lozoya R, Ziupa D, Menza M, Relan J, Franke G, Perez-Feliz S, Koren G, Zehender M, Bode C, Brunner M, Sermesant M, Föll D. Spatial correlation of action potential duration and diastolic dysfunction in transgenic and drug-induced LQT2 rabbits. *Heart Rhythm* 2013;**10**:1533–1541.
114. Ziv O, Morales E, Song YK, Peng X, Odening KE, Buxton AE, Karma A, Koren G, Choi BR. Origin of complex behaviour of spatially discordant alternans in a transgenic rabbit model of type 2 long QT syndrome. *J Physiol* 2009;**587**:4661–4680.
115. Ziupa D, Beck J, Franke G, Perez-Feliz S, Hartmann M, Koren G, Zehender M, Bode C, Brunner M, Odening KE. Pronounced effects of HERG-blockers E-4031 and erythromycin on APD, spatial APD dispersion and triangulation in transgenic long-QT type 1 rabbits. *PLoS One* 2014;**9**:e107210.
116. Lau E, Kossidas K, Kim TY, Kunitomo Y, Ziv O, Song Z, Taylor C, Schofield L, Yammine J, Liu G, Peng X, Qu Z, Koren G, Choi BR. Spatially discordant alternans and arrhythmias in tachypacing-induced cardiac myopathy in transgenic LQT1 rabbits: the importance of I_{Ks} and Ca²⁺ cycling. *PLoS One* 2015;**10**:e0122754.
117. Kim TY, Kunitomo Y, Pfeiffer Z, Patel D, Hwang J, Harrison K, Patel B, Peng P, Ziv O, Lu Y, Peng X, Qu Z, Koren G, Choi BR. Complex excitation dynamics underlie polymorphic ventricular tachycardia in a transgenic rabbit model of long QT syndrome type 1. *Heart Rhythm* 2015;**12**:220–228.
118. Odening KE, Hyder O, Chaves L, Schofield L, Brunner M, Kirk M, Zehender M, Peng X, Koren G. Pharmacogenomics of anesthetic drugs in transgenic LQT1 and LQT2 rabbits reveal genotype-specific differential effects on cardiac repolarization. *Am J Physiol Heart Circ Physiol* 2008;**295**:H2264–H2272.
119. Bodi I, Grunert SC, Becker N, Stoelele-Feix S, Spiekerkoetter U, Zehender M, Bugger H, Bode C, Odening KE. Mechanisms of acquired long QT syndrome in patients with propionic academia. *Heart Rhythm* 2016;**13**:1335–1345.
120. Bodi I, Sorge J, Castiglione A, Glatz SM, Wuelfers EM, Franke G, Perez-Feliz S, Koren G, Zehender M, Bugger H, Seemann G, Brunner M, Bode C, Odening KE. Postpartum hormones oxytocin and prolactin cause pro-arrhythmic prolongation of cardiac repolarization in long QT syndrome type 2. *Europace* 2019;**21**:1126–1138.
121. Lang CN, Menza M, Jochem S, Franke G, Perez-Feliz S, Brunner M, Koren G, Zehender M, Bugger H, Jung BA, Foell D, Bode C, Odening KE. Electro-mechanical dysfunction in long QT syndrome: role for arrhythmogenic risk prediction and modulation by sex and sex hormones. *Prog Biophys Mol Biol* 2016;**120**:255–269.
122. Milan DJ, Macrae CA. Zebrafish genetic models for arrhythmia. *Prog Biophys Mol Biol* 2008;**98**:301–308.
123. Nemtsas P, Wettwer E, Christ T, Weidinger G, Ravens U. Adult zebrafish heart as a model for human heart? An electrophysiological study. *J Mol Cell Cardiol* 2010;**48**:161–171.
124. Ravens U. Ionic basis of cardiac electrophysiology in zebrafish compared to human hearts. *Prog Biophys Mol Biol* 2018;**138**:38–44.
125. Langheinrich U, Vacun G, Wagner T. Zebrafish embryos express an orthologue of HERG and are sensitive toward a range of QT-prolonging drugs inducing severe arrhythmia. *Toxicol Appl Pharmacol* 2003;**193**:370–382.
126. McGrath P, Li CQ. Zebrafish: a predictive model for assessing drug-induced toxicity. *Drug Discov Today* 2008;**13**:394–401.
127. Arnaout R, Ferrer T, Huisken J, Spitzer K, Stainier DY, Tristani-Firouzi M, Chi NC. Zebrafish model for human long QT syndrome. *Proc Natl Acad Sci USA* 2007;**104**:11316–11321.
128. Peal DS, Mills RW, Lynch SN, Mosley JM, Lim E, Ellinor PT, January CT, Peterson RT, Milan DJ. Novel chemical suppressors of long QT syndrome identified by an in vivo functional screen. *Circulation* 2011;**123**:23–30.
129. Milan DJ, Kim AM, Winterfield JR, Jones IL, Pfeufer A, Sanna S, Arking DE, Amsterdam AH, Sabeh KM, Mably JD, Rosenbaum DS, Peterson RT, Chakravarti A, Käb S, Roden DM, MacRae CA. Drug-sensitized zebrafish screen identifies multiple genes, including GINS3, as regulators of myocardial repolarization. *Circulation* 2009;**120**:553–559.
130. Moretti A, Bellin M, Welling A, Jung CB, Lam JT, Bott-Flügel L, Dorn T, Goedel A, Höhnke F, Hofmann F, Seyfarth M, Sinnecker D, Schömig A, Laugwitz K-L. Patient-specific induced pluripotent stem-cell models for long-QT syndrome. *N Engl J Med* 2010;**363**:1397–1409.
131. Itzhaki I, Maizels L, Huber I, Zwi-Dantsis L, Caspi O, Winterstern A, Feldman O, Gepstein A, Arbel G, Hammerman H, Boulos M, Gepstein L. Modelling the long QT syndrome with induced pluripotent stem cells. *Nature* 2011;**471**:225–229.
132. Malan D, Friedrichs S, Fleischmann BK, Sasse P. Cardiomyocytes obtained from induced pluripotent stem cells with long-QT syndrome 3 recapitulate typical disease-specific features in vitro. *Circ Res* 2011;**109**:841–847.
133. Mehta A, Ramachandra CJA, Singh P, Chitre A, Lua CH, Mura M, Crotti L, Wong P, Schwartz PJ, Gnechi M, Shim W. Identification of a targeted and testable antiarrhythmic therapy for long-QT syndrome type 2 using a patient-specific cellular model. *Eur Heart J* 2018;**39**:1446–1455.
134. Schwartz PJ, Gnechi M, Dagradi F, Castelletti S, Parati G, Spazzolini C, Sala L, Crotti L. From patient-specific induced pluripotent stem cells to clinical translation in long QT syndrome Type 2. *Eur Heart J* 2019;**40**:1832–1836.
135. Robertson GA, January CT. HERG trafficking and pharmacological rescue of LQTS-2 mutant channels. *Handb Exp Pharmacol* 2006;**171**:349–355.
136. Matsa E, Dixon JE, Medway C, Georgiou O, Patel MJ, Morgan K, Kemp PJ, Staniforth A, Mellor I, Denning C. Allele-specific RNA interference rescues the long-QT syndrome phenotype in human-induced pluripotency stem cell cardiomyocytes. *Eur Heart J* 2014;**35**:1078–1087.
137. Mattmann ME, Yu H, Lin Z, Xu K, Huang X, Long S, Wu M, McManus OB, Engers DW, Le UM, Li M, Lindsley CW, Hopkins CR. Identification of (R)-N-(4-(4-methoxyphenyl)thiazol-2-yl)-1-tosylpiperidine-2-carboxamide, ML277, as a novel, potent and selective Kv7.1 (KCNQ1) potassium channel activator. *Bioorg Med Chem Lett* 2012;**22**:5936–5941.
138. Hou P, Shi J, White KM, Gao Y, Cui J. ML277 specifically enhances the fully activated open state of KCNQ1 by modulating VSD-pore coupling. *Elife* 2019;**8**:e48576.
139. Ma D, Wei H, Lu J, Huang D, Liu Z, Loh LJ, Islam O, Liew R, Shim W, Cook SA. Characterization of a novel KCNQ1 mutation for type 1 long QT syndrome and assessment of the therapeutic potential of a novel IKs activator using patient-specific induced pluripotent stem cell-derived cardiomyocytes. *Stem Cell Res Ther* 2015;**6**:39.
140. Kang J, Chen XL, Wang H, Ji J, Cheng H, Incardona J, Reynolds W, Viviani F, Tabart M, Rampe D. Discovery of a small molecule activator of the human ether- α -go-go-related gene (HERG) cardiac K⁺ channel. *Mol Pharmacol* 2005;**67**:827–836.
141. Gerlach AC, Stoehr SJ, Castle NA. Pharmacological removal of human ether- α -go-go-related gene potassium channel inactivation by 3-nitro-N-(4-phenoxyphenyl) benzamide (ICA-105574). *Mol Pharmacol* 2010;**77**:58–68.
142. Zhang H, Zou B, Yu H, Moretti A, Wang X, Yan W, Babcock JJ, Bellin M, McManus OB, Tomaselli G, Nan F, Laugwitz KL, Li M. Modulation of HERG potassium channel gating normalizes action potential duration prolonged by dysfunctional KCNQ1 potassium channel. *Proc Natl Acad Sci USA* 2012;**109**:11866–11871.
143. Mannikko R, Bridgland-Taylor MH, Pye H, Swallow S, Abi-Gerges N, Morton MJ, Pollard CE. Pharmacological and electrophysiological characterization of AZSMO-23, an activator of the HERG K⁺ channel. *Br J Pharmacol* 2015;**172**:3112–3125.

144. Perry M, Sachse FB, Abbruzzese J, Sanguinetti MC. PD-118057 contacts the pore helix of hERG1 channels to attenuate inactivation and enhance K⁺ conductance. *Proc Natl Acad Sci USA* 2009;**106**:20075–20080.
145. Hansen RS, Dinness TG, Christ T, Demnitz J, Ravens U, Olesen SP, Grunnet M. Activation of human ether-a-go-go-related gene potassium channels by the diphenylurea 1,3-bis-(2-hydroxy-5-trifluoromethyl-phenyl)-urea (NS1643). *Mol Pharmacol* 2006;**69**:266–277.
146. Bentzen BH, Bahrke S, Wu K, Larsen AP, Odening KE, Franke G, varS Gravesande KS, Biermann J, Peng X, Koren G, Zehender M, Bode C, Grunnet M, Brunner M. Pharmacological activation of Kv11.1 in transgenic long QT-1 rabbits. *J Cardiovasc Pharmacol* 2011;**57**:223–230.
147. Giustetto C, Di Monte F, Wolpert C, Borggreffe M, Schimpf R, Sbragia P, Leone G, Maury P, Anttonen O, Haissaguerre M, Gaita F. Short QT syndrome: clinical findings and diagnostic-therapeutic implications. *Eur Heart J* 2006;**27**:2440–2447.
148. Mazzanti A, Kanthan A, Monteforte N, Memmi M, Bloise R, Novelli V, Miceli C, O'Rourke S, Borio G, Zienciu-Krajka A, Curcio A, Surducian AE, Colombo M, Napolitano C, Priori SG. Novel insight into the natural history of short QT syndrome. *J Am Coll Cardiol* 2014;**63**:1300–1308.
149. Rhodes TE, Abraham RL, Welch RC, Vanoye CG, Crotti L, Arnestad M, Insolia R, Pedrazzini M, Ferrandi C, Vege A, Rognum T, Roden DM, Schwartz PJ, George AL Jr. Cardiac potassium channel dysfunction in sudden infant death syndrome. *J Mol Cell Cardiol* 2008;**44**:571–581.
150. El-Battrawy I, Schlenkrich K, Besler J, Liebe V, Schimpf R, Lang S, Odening KE, Wolpert C, Zhou X, Borggreffe M, Akin I. Sex-differences in short QT syndrome: a systematic literature review and pooled analysis. *Eur J Prev Cardiol* 2019;doi: 10.1177/2047487319850953.
151. Bjerregaard P. Proposed diagnostic criteria for short QT syndrome are badly founded. *J Am Coll Cardiol* 2011;**58**:549–550.
152. El-Battrawy I, Besler J, Liebe V, Schimpf R, Tulumen E, Rudic B, Lang S, Wolpert C, Zhou X, Akin I, Borggreffe M. Long-term follow-up of patients with short QT syndrome: clinical profile and outcome. *J Am Heart Assoc* 2018;**7**:e010073.
153. Harrell DT, Ashihara T, Ishikawa T, Tominaga I, Mazzanti A, Takahashi K, Oginozawa Y, Abe H, Maemura K, Sumitomo N, Uno K, Takano M, Priori SG, Makita N. Genotype-dependent differences in age of manifestation and arrhythmia complications in short QT syndrome. *Int J Cardiol* 2015;**190**:393–402.
154. Hong K, Piper DR, Diaz-Valdecantos A, Brugada J, Oliva A, Burashnikov E, Santos-de-Soto J, Gruesso-Montero J, Diaz-Enfante E, Brugada P, Sachse F, Sanguinetti MC, Brugada R. De novo KCNQ1 mutation responsible for atrial fibrillation and short QT syndrome in utero. *Cardiovasc Res* 2005;**68**:433–440.
155. Gussak I, Brugada P, Brugada J, Wright RS, Kopecky SL, Chaitman BR, Bjerregaard P. Idiopathic short QT interval: a new clinical syndrome? *Cardiology* 2000;**94**:99–102.
156. Brugada R, Hong K, Dumaine R, Cordeiro J, Gaita F, Borggreffe M, Menendez TM, Brugada J, Pollevick GD, Wolpert C, Burashnikov E, Matsuo K, Wu YS, Guerschicoff A, Bianchi F, Giustetto C, Schimpf R, Brugada P, Antzelevitch C. Sudden death associated with short-QT syndrome linked to mutations in HERG. *Circulation* 2004;**109**:30–35.
157. Bellocq C, van Ginneken AC, Bezzina CR, Alders M, Escande D, Mannens MM, Baro I, Wilde AA. Mutation in the KCNQ1 gene leading to the short QT-interval syndrome. *Circulation* 2004;**109**:2394–2397.
158. Priori SG, Pandit SV, Rivolta I, Berenfeld O, Ronchetti E, Dharmoon A, Napolitano C, Anumonwo J, di Barletta MR, Gudapakkam S, Bosi G, Stramba-Badiale M, Jalife J. A novel form of short QT syndrome (SQT3) is caused by a mutation in the KCNJ2 gene. *Circ Res* 2005;**96**:800–807.
159. Sun Y, Quan XQ, Fromme S, Cox RH, Zhang P, Zhang L, Guo D, Guo J, Patel C, Kowey PR, Yan GX. A novel mutation in the KCNH2 gene associated with short QT syndrome. *J Mol Cell Cardiol* 2011;**50**:433–441.
160. Peng G, Barro-Soria R, Sampson KJ, Larsson HP, Kass RS. Gating mechanisms underlying deactivation slowing by two KCNQ1 atrial fibrillation mutations. *Sci Rep* 2017;**7**:45911.
161. Deo M, Ruan Y, Pandit SV, Shah K, Berenfeld O, Blafox A, Cerrone M, Noujaim SF, Denegri M, Jalife J, Priori SG. KCNJ2 mutation in short QT syndrome 3 results in atrial fibrillation and ventricular proarrhythmia. *Proc Natl Acad Sci USA* 2013;**110**:4291–4296.
162. Ambrosini E, Sicca F, Brignone MS, D'Adamo MC, Napolitano C, Servetini I, Moro F, Ruan Y, Guglielmi L, Pieroni S, Servillo G, Lanciotti A, Valvo G, Catacuzzano L, Franciolini F, Molinari P, Marchese M, Grottesi A, Guerrini R, Santorelli FM, Priori S, Pessia M. Genetically induced dysfunctions of Kir2.1 channels: implications for short QT3 syndrome and autism-epilepsy phenotype. *Hum Mol Genet* 2014;**23**:4875–4886.
163. Priori SG, Blomström-Lundqvist C, Mazzanti A, Blom N, Borggreffe M, Camm J, Elliott PM, Fitzsimons D, Hatala R, Hindricks G, Kirchhof P, Kjeldsen K, Kuck K-H, Hernandez-Madrid A, Nikolaou N, Norekvål TM, Spaulding C, Van Veldhuisen DJ; ESC Scientific Document Group. 2015 ESC Guidelines for the management of patients with ventricular arrhythmias and the prevention of sudden cardiac death: the Task Force for the Management of Patients with Ventricular Arrhythmias and the Prevention of Sudden Cardiac Death of the European Society of Cardiology (ESC). Endorsed by: association for European Paediatric and Congenital Cardiology (AEPC). *Europace* 2015;**17**:1601–1687.
164. Giustetto C, Scrocco C, Schimpf R, Maury P, Mazzanti A, Levetto M, Anttonen O, Dalmasso P, Cerrato N, Gribaudo E, Wolpert C, Giachino D, Antzelevitch C, Borggreffe M, Gaita F. Usefulness of exercise test in the diagnosis of short QT syndrome. *Europace* 2015;**17**:628–634.
165. Anttonen O, Junttila MJ, Maury P, Schimpf R, Wolpert C, Borggreffe M, Giustetto C, Gaita F, Sacher F, Haissaguerre M, Sbragia P, Brugada R, Huikuri HV. Differences in twelve-lead electrocardiogram between symptomatic and asymptomatic subjects with short QT interval. *Heart Rhythm* 2009;**6**:267–271.
166. Borggreffe M, Wolpert C, Antzelevitch C, Veltmann C, Giustetto C, Gaita F, Schimpf R. Short QT syndrome. Genotype-phenotype correlations. *J Electrocardiol* 2005;**38**:75–80.
167. Gussak I, Antzelevitch C. Early repolarization syndrome: clinical characteristics and possible cellular and ionic mechanisms. *J Electrocardiol* 2000;**33**:299–309.
168. Hassel D, Scholz EP, Trano N, Friedlich O, Just S, Meder B, Weiss DL, Zitron E, Marquart S, Vogel B, Karle CA, Seemann G, Fishman MC, Katus HA, Rottbauer W. Deficient zebrafish ether-a-go-go-related gene channel gating causes short-QT syndrome in zebrafish reggae mutants. *Circulation* 2008;**117**:866–875.
169. Odening KE, Bodi I, Franke G, Rieke R, Ryan de Medeiros A, Perez-Feliz S, Furniss H, Mettke L, Michaelides K, Lang CN, Steinfurt J, Pantulu ND, Ziupa D, Menza M, Zehender M, Bugger H, Peyronnet R, Behrends JC, Doleschall Z, Zur Hausen A, Bode C, Jolivet G, Brunner M. Transgenic short-QT syndrome 1 rabbits mimic the human disease phenotype with QT/action potential duration shortening in the atria and ventricles and increased ventricular tachycardia/ventricular fibrillation inducibility. *Eur Heart J* 2019;**40**:842–853.
170. Shinnawi R, Shaheen N, Huber I, Shiti A, Arbel G, Gepstein A, Ballan N, Setter N, Tijssen AJ, Borggreffe M, Gepstein L. Modeling reentry in the short QT syndrome with human-induced pluripotent stem cell-derived cardiac cell sheets. *J Am Coll Cardiol* 2019;**73**:2310–2324.
171. Guo F, Sun Y, Wang X, Wang H, Wang J, Gong T, Chen X, Zhang P, Su L, Fu G, Su J, Yang S, Lai R, Jiang C, Liang P. Patient-specific and gene-corrected induced pluripotent stem cell-derived cardiomyocytes elucidate single-cell phenotype of short QT syndrome. *Circ Res* 2019;**124**:66–78.
172. El-Battrawy I, Lan H, Cyganek L, Zhao Z, Li X, Buljubasic F, Lang S, Yücel G, Sattler K, Zimmermann WH, Utikal J, Wieland T, Ravens U, Borggreffe M, Zhou XB, Akin I. Modeling short QT syndrome using human-induced pluripotent stem cell-derived cardiomyocytes. *J Am Heart Assoc* 2018;**7**:e007394.
173. Priori SG, Wilde AA, Horie M, Cho Y, Behr ER, Berul C, Blom N, Brugada J, Chiang CE, Huikuri H, Kannankeril P, Krahn A, Leenhardt A, Moss A, Schwartz PJ, Shimizu W, Tomaselli G, Tracy C. Executive summary: HRS/EHRA/APHRS expert consensus statement on the diagnosis and management of patients with inherited primary arrhythmia syndromes. *Europace* 2013;**15**:1389–1406.
174. Schimpf R, Wolpert C, Bianchi F, Giustetto C, Gaita F, Bauersfeld U, Borggreffe M. Congenital short QT syndrome and implantable cardioverter defibrillator treatment: inherent risk for inappropriate shock delivery. *J Cardiovasc Electrophysiol* 2003;**14**:1273–1277.
175. El-Battrawy I, Besler J, Ansari U, Liebe V, Schimpf R, Tulumen E, Rudic B, Lang S, Odening K, Cyganek L, Wolpert C, Zhou X, Borggreffe M, Akin I. Long-term follow-up of implantable cardioverter-defibrillators in short QT syndrome. *Clin Res Cardiol* 2019;**108**:1140–1146.
176. Mazzanti A, Maragna R, Vacanti G, Kostopoulou A, Marino M, Monteforte N, Bloise R, Underwood K, Tibollo V, Pagan E, Napolitano C, Bellazzi R, Bagnardi V, Priori SG. Hydroquinidine prevents life-threatening arrhythmic events in patients with short QT syndrome. *J Am Coll Cardiol* 2017;**70**:3010–3015.
177. Giustetto C, Scrocco C, Giachino D, Rapezzi C, Mognetti B, Gaita F. The lack of effect of sotalol in short QT syndrome patients carrying the T618I mutation in the KCNH2 gene. *HeartRhythm Case Rep* 2015;**1**:373–378.
178. Hu D, Li Y, Zhang J, Pfeiffer R, Gollob MH, Healey J, Harrell DT, Makita N, Abe H, Sun Y, Guo J, Zhang L, Yan G, Mah D, Walsh EP, Leopold HB, Giustetto C, Gaita F, Zienciu-Krajka A, Mazzanti A, Priori SG, Antzelevitch C, Barajas-Martinez H. The phenotypic spectrum of a mutation hotspot responsible for the short QT syndrome. *JACC Clin Electrophysiol* 2017;**3**:727–743.
179. El Harchi A, McPate MJ, Zhang Y, Zhang H, Hancox JC. Action potential clamp and chloroquine sensitivity of mutant Kir2.1 channels responsible for variant 3 short QT syndrome. *J Mol Cell Cardiol* 2009;**47**:743–747.
180. Hancox JC, Whittaker DG, Du C, Stuart AG, Zhang H. Emerging therapeutic targets in the short QT syndrome. *Expert Opin Ther Targets* 2018;**22**:439–451.