Cardiac sodium channel and inherited arrhythmia syndromes

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1. Introduction

The voltage-gated cardiac sodium channel is responsible for the generation of the rapid upstroke of the myocardial action potential and thereby plays a central role in excitability of myocardial cells. In addition, since the action potential upstroke velocity — in conjunction with the extent of intercellular communication via gap junctions [1] — also determines impulse conduction velocity in cardiac tissue, this channel also plays a vital role in impulse propagation. Mutations in the gene encoding this channel (\textit{SCN5A}; Section 2.1) have been linked to three forms of primary electrical disease — the long QT syndrome (LQTS) [2], the Brugada syndrome (BS) [3] and cardiac conduction defects [4]. The elucidation of the pathophysiological mechanisms of these mutant Na\textsuperscript{+} channels would ultimately enable more specific pharmacological intervention in the management of these syndromes and other related arrhythmias.

2. Cardiac sodium channel

The cardiac Na\textsuperscript{+} channel (see Ref. [5] for review) is a member of the voltage-dependent family of Na\textsuperscript{+} channels (see Ref. [6] for review). These channels consist of heteromeric assemblies of an \(\alpha\)-subunit, the pore-forming component, the function of which is modulated by association with one or two ancillary \(\beta\)-subunits.

2.1. \(\alpha\)-Subunit

The human cardiac Na\textsuperscript{+} channel \(\alpha\)-subunit is a heavily glycosylated protein of \(\sim 260\) kDa consisting of 2016 amino acid residues [7]. It is encoded by the \textit{SCN5A} gene [8], which is located on chromosome 3p21 [9]. At least 11 other different genes that encode highly homologous sodium channel \(\alpha\)-subunit isoforms expressed in the heart, muscle and nervous system have been hitherto identified in man (http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?db =Nucleotide). They display a modular architecture (Fig. 1), which consists of four internally homologous domains (DI–DIV) each made up of six transmembrane segments (S1–S6). The interdomain linkers and the N- and C-terminal ends of the channel protein are all located cytoplasmically.

Immunocytochemical experiments conducted in adult rat heart tissue revealed that in addition to the expected localization of Na\textsuperscript{+} channels at the surface and t-tubular membranes, Na\textsuperscript{+} channels localize to terminal intercalated disks [10], also the predominant sites of gap junctional coupling. It has been proposed that this subcellular localization could play a role in anisotropic or saltatory conduction, or may act as a localized voltage-dependent current amplifier [10].

2.1.1. Permeation

The four domains of the channel fold around a central ion-conducting pore [11], which is lined by the S5–S6 linker (referred to as the P-segment or P-loop) from each domain [12]. This loop, which exhibits a high degree of conservation among the various organ-specific isoforms across species, determines selectivity and conductance properties of the channel [13]. Each of the four P-loop domains has a unique primary structure and a different positioning relative to the permeation pathway [14–16]. Thus, the P-loop from each domain exerts a unique contribution towards the ion selectivity, ion and toxin

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binding properties and permeation characteristics of the channel. In particular, the P-loops of domains III and IV play an important role in Na\(^+\) selectivity. A lysine residue in the P-loop of DIII (K1418 in SCN5A), is critical for discrimination for Na\(^+\) over Ca\(^{2+}\) [13,17], and, in DIV, mutations of the tetrad WDGL (residues 1713–1716 in SCN5A) affect selectivity among monovalent cations [14]. In line with the analogy of ion channels as enzymes — catalysing selective passage of ions across the cell membrane [18] — it has been shown that P-loops are remarkably flexible structures [19,20] and that this flexibility could enhance selective ion translocation [19].

The cardiac Na\(^+\) channel differs from the neuronal and skeletal muscle isoforms in its sensitivity to block by tetrodotoxin (TTX). Whereas brain and skeletal muscle isoforms are blocked at nanomolar concentrations of TTX, the heart isoform is blocked at micromolar concentrations. Residues in the P-loop of DI are responsible for this isoform-specific difference in toxin binding. A cysteine at position 373 within the heart isoform is responsible for the relative TTX resistance of this isoform, whereas the presence of aromatic residues (tyrosine or phenylalanine) at the corresponding position in the skeletal muscle and neuronal isoforms renders them TTX sensitive [21]. This single amino acid difference is also responsible for the susceptibility of the cardiac isoform to Cd\(^{2+}\) block, compared to the insensitivity of the neuronal and skeletal muscle isoforms to block by this divalent cation [22].

2.1.2. Gating

Na\(^+\) channels transit among various conformational states in the process of voltage-dependent gating. Depolarization from the resting membrane potential triggers activation (opening) of the Na\(^+\) channels. If the depolarization is maintained, the channels enter a non-conducting inactivated state. Subsequent to repolarization, the channels return to a closed state capable of being activated once again. It is becoming increasingly clear that these processes are the result of complex allosteric interactions among many structural domains of the channel.

The fourth transmembrane segment in each domain (S4, Fig. 1) functions as a voltage sensor and is responsible for activation gating [23]. It contains positively charged residues at every third position (arginine or lysine), which bring about motion of S4 segments in response to depolarization [24–26], leading to opening of the channel pore. All four S4 segments contribute to voltage-dependent activation of the Na\(^+\) channel; however, differentiation in function between the four domains is also evidenced here. Positively charged residues at comparable positions within the different S4 segments contribute unequally towards the voltage-dependent properties of the channel [27]. The structure(s) that forms the actual gates is yet unknown, but is expected to lie on the cytoplasmic side of the channel [6].

Inactivation is characterized by at least two distinguishable kinetic components, an initial rapid component with a
fast recovery time constant (fast inactivation) and a slower component with a slower recovery time constant (slow inactivation). Fast inactivation is mediated, at least in part, by the DIII–DIV cytoplasmic linker (Fig. 1) [23,28]. This linker can be visualized as a hinged lid, which docks against receptor sites surrounding the inner vestibule of the pore, thereby occluding the pore. A critical component of DIII–DIV required for fast inactivation has been localized to the highly conserved hydrophobic triad IFM (residues 1485–1487 in the cardiac isoform) [29]. These residues are thought to interact with a receptor site that becomes available in the activated Na\(^+\) channel [29,30]. The S4–S5 linkers in domains III and IV [31–35] and residues on the cytoplasmic end of S6 of DIV [36] could form part of this receptor site. It has also been proposed that rather than being involved in direct interaction with the IFM motif itself, S6 could be critical for fast inactivation by stabilizing the inactivation state through an indirect mechanism [37].

Structural determinants of slow inactivation are less known but are most probably localized within P-loops. It has been demonstrated that slow inactivation can be modulated by mutation of elements within the P-loops [38–42], and that transposition of all four human heart isoform P-loops into the human skeletal muscle isoform (hSKM1) backbone conferred heart isoform-like slow inactivation properties on the chimeric construct [43]. This suggests that slow inactivation in Na\(^+\) channels involves conformational changes in the outer pore in a fashion analogous to C-type inactivation in K\(^+\) channels [44,45]. Flexibility of the P-loops [19,20] could affect this process [46].

Inactivation derives most of its voltage dependence from being coupled to activation [47]. This implies that the rate of inactivation increases as a consequence of conformational changes in the channel associated with activation. The molecular details underlying the coupling of inactivation to activation are starting to be unravelled. It appears that the voltage sensing S4 segments, at least of DIV, play a crucial role in this process [48–54,24,35]. Movement of the voltage sensors in the activation pathway possibly brings about a reorganisation of the docking site for the inactivation gate, thereby easing binding of the inactivation gate. Another mechanism based on a physical interaction between regions on the S4–S5 linker of DIV and the DIII–DIV inactivation particle has also been proposed [55]. It has been suggested that such an association could be sensitive to changes in the conformation of S4–S5 of DIV, which are brought about by movement of the voltage sensor in that domain during activation. This would result in a change in the position of the inactivation gate relative to its docking sites.

2.2. \(\beta\)-1-Subunit

Auxiliary (\(\beta\)) subunits have been shown to be important modulators of Na\(^+\) channels from brain and skeletal muscle. The brain Na\(^+\) channel consists of the \(\alpha\)-subunit, a non-covalently bound \(\beta\)-1-subunit and a disulfide-linked \(\beta\)-2-subunit [56,57]. The skeletal muscle Na\(^+\) channel consists of the \(\alpha\)-subunit and a non-covalently linked \(\beta\)-1-subunit [58,59]. These \(\beta\)-1 and \(\beta\)-2-subunits are unrelated proteins of 36 and 33 kDa, respectively, each consisting of a small C-terminal cytoplasmic domain, a single transmembrane segment, and a large glycosylated N-terminal extracellular domain [60–62].

The human \(\beta\)-1-subunit is a 218-amino-acid protein [63] encoded by the SCN1B gene located on chromosome 19q13.1 [64]. It is highly expressed in the heart, skeletal muscle and brain [62]. Co-expression of brain or skeletal Na\(^+\) channel \(\alpha\)-subunit isoforms with the \(\beta\)-1-subunit in Xenopus oocytes have consistently demonstrated a larger peak \(I_{\text{m}}\) amplitude, acceleration of activation and inactivation, and a more negative voltage dependence of steady-state inactivation than the \(\alpha\)-subunit expressed alone [60,62,63,65–70], demonstrating that co-expression with the \(\beta\)-1-subunit recapitulates the characteristics of the channels observed in vivo by modulating their gating and increasing the efficiency of their expression.

Contrary to the clear role of the \(\beta\)-1-subunit in skeletal muscle and the nervous system, the occurrence of such an interaction with the cardiac \(\alpha\)-subunit isoform in vivo is not clear. Studies using subunit-specific antibodies have identified the \(\beta\)-1-subunit polypeptide in the heart [71]. \(\beta\)-1-subunit mRNA has also been detected by Northern blot analyses [60,67,68,62], in situ hybridization [72] as well as RT-PCR on RNA from isolated ventricular myocytes [73]. However, immunoprecipitation experiments suggest that the cardiac \(\alpha\)-subunit isoform is not associated with a \(\beta\)-subunit [74].

Moreover, co-expression studies of the cardiac Na\(^+\) channel \(\alpha\)-subunit with the \(\beta\)-1-subunit in Xenopus oocytes have produced conflicting results. Some investigators [62,68] reported no effect of the \(\beta\)-1-subunit on the kinetics of the cardiac Na\(^+\) channel \(\alpha\)-subunit. Others [70,72] have reported little or no effect on the kinetics or voltage dependence of the Na\(^+\) current but showed a substantial increase in Na\(^+\) channel expression manifested as an increase in peak current amplitude. Another group [73] reported a 3–7 mV shift in the depolarizing direction in the voltage dependence of inactivation upon co-expression of the \(\beta\)-1-subunit. Wei et al. [75] also reported a 10-mV shift in the depolarizing direction upon co-expression with the \(\beta\)-1-subunit. A more recent study in human embryonic kidney (HEK 293) cells has also shown a shift (11 mV) in the depolarising direction upon co-expression of the \(\alpha\)- and \(\beta\)-1-subunits compared to expression of the \(\alpha\)-subunit alone [76]. Suppression of \(\beta\)-1 expression in mouse atrial tumor (AT-1) cells by antisense oligonucleotides resulted in a reduction in \(I_{\text{m}}\) amplitude, slower activation and inactivation, and a less negative voltage dependence of inactivation [77], effects which are in accordance with those observed for \(\beta\)-1 on the brain and skeletal muscle \(\alpha\)-subunits. This data, together with the fact that transcription
of the β1-subunit increases dramatically postnatally [77], suggest that the α–β1 interaction underlies the developmental change of $I_{Na}$ from the neonatal phenotype to the adult phenotype [77] which is characterized by much more rapid activation and inactivation and a more negative midpoint for inactivation [78]. Development of a mature $I_{Na}$ is also promoted by an intracellular signalling event [78–80]. However, the mechanism explaining the relationship between intracellular signalling and α–β1 interaction still needs to be worked out. The developmental phenotypic change in $I_{Na}$ could underlie the increase in maximum upstroke velocity and conduction velocity observed with development [81].

Expression studies on two naturally occurring Na$^+$ channel mutants associated with LQTS type 3 (LQTS$_3$) and the BS, respectively, have shown that their functional defect was accentuated by co-expression of the β1-subunit (Sections 3.1 and 4.2; Tables 1 and 2), hinting at an interaction in vivo. On the other hand, mutations in SCN1B have so far been linked only to febrile seizures and generalized epilepsy [82].

Structural correlates for the α–β1 interaction are thought to reside on the extracellular and intramembraneous domains of the β1-subunit [83–85] and P-loops within the α-subunit [86,43]. The cytoplasmic C-terminal domain of the α-subunit could also play a role [76]. A role for the β1-subunit in Na$^+$ channel localization has been proposed on the basis of the inferred immunoglobulin-like structure of its large extracellular domain [87].

2.3. Second messenger regulation

Signalling pathways that may play a role in regulation of cardiac Na$^+$ channel function include those leading to activation of cAMP-dependent protein kinase (protein kinase A, PKA) and protein kinase C (PKC). Modulation by PKA occurs via phosphorylation at two serine residues (S525 and S528 in the human cardiac isoform) within the DI–DII linker [88]. Phosphorylation by PKA at these sites augments the Na$^+$ current [88–90]. However, there have been conflicting findings relating to the cardiac Na$^+$ current modulation by β-adrenergic stimulation, with studies showing enhancement of $I_{Na}$ [91–93] and others showing inhibition of $I_{Na}$ by isoproterenol [94–96]. However, one should note that the different investigators performed experiments on myocytes from different species, at different stages of development and possibly also from different parts of the heart.

Moreover, in addition to the indirect pathway mediated by G-proteins acting through second messenger cascades, a direct membrane-delimited G-protein regulatory pathway involving direct interaction of the stimulatory G-protein α-subunit with the cardiac Na$^+$ channel has also been put forward [91,93,94].

Modulation by PKC occurs via phosphorylation at S1505 in the DIII–DIV inactivation gate [97]. Phosphorylation at this site results in a negative shift in the voltage-dependence of steady-state inactivation and a reduction in current amplitude [97].

3. Long QT syndrome

The LQTS (Ref. [98] for review) is an inherited cardiac arrhythmia that may cause abrupt syncope, seizures and sudden death from ventricular tachyarrhythmias, specifically torsades de pointes, and ventricular fibrillation. In addition to the prolonged QT-interval, electrocardiographic abnormalities include T-wave abnormalities such as notched or biphasic T waves, T-wave alternans, prominent U waves and bradycardia. This syndrome arises from delayed repolarization of cardiomyocytes resulting from an imbalance in the inward and outward currents during the repolarization phase of the myocardial action potential. It has been estimated to affect 1 in 5000 individuals.

LQTS is often transmitted in families as an autosomal dominant trait and less often as an autosomal recessive trait combined with congenital sensorineural deafness. In some families a very low penetrance for the disorder has been described [99].

One subtype of this disorder — LQTS$_1$ — is caused by mutations in SCN5A [2] (Section 3.1). The other forms are caused by mutations in genes encoding subunits of repolarizing delayed rectifier potassium channels, namely KCNQ1, KCNH2, KCNE1, and KCNE2. An as yet unidentified gene for this disorder has been mapped to chromosome 4 [98].

3.1. SCN5A mutations in LQTS$_3$

Mutations in SCN5A were linked to LQTS$_3$ by a positional candidate gene approach. Jiang et al. [100] mapped the LQTS$_3$ locus to 3p21–24. SCN5A was soon after mapped to chromosome 3p21 [9], making it an excellent candidate gene for LQTS$_3$. In the same year, Wang et al. [2] reported an identical SCN5A mutation — deletion of residues K1505, P1506, Q1507 (ΔKPQ) — in affected members of two families.

Nine distinct mutations associated with LQTS$_3$ (Fig. 1, Table 1), all located in domains III and IV, have been reported to date. Apart from the ΔKPQ mutation [2], they involve amino acid substitutions (S941N [101]; N1325S, R1644H [102]; R1623Q [103]; D1790G [104]; E1784K [75,105]; T1304M, T1645M [106]). The biophysical properties of almost all LQTS$_3$-related mutant channels (all except for T1304M and T1645M channels) have been investigated by expression in Xenopus oocytes or mammalian cell lines (Table 1). All are associated with the occurrence of Na$^+$ channels with altered properties. Heterologous expression of ΔKPQ, N1325S and R1644H mutant channels [32,107,108] demonstrated that these channels display macroscopic current decay rates comparable to the wild-type channel, or may even have faster decay rates (Table 1, ΔKPQ mutant), but exhibit small persistent inward currents resulting from channel...
The late-opening behaviour is more extensive in the case of the ΔKPQ mutant. The ΔKPQ mutation is located in the inactivation gate (DIII–DIV linker involved in fast inactivation) of the channel while the N1325S and R1644H mutations are located on the intracellular surface at sites that could constitute docking sites for the inactivation gate associated with a small persistent inward current, and a T917K mutation is located in the C-terminal domain, immediately following the DIVS6 segment of the channel protein leads to a persistent inward current. The mechanism whereby substitution within this part of the channel protein leads to a persistent inward current is yet unclear. R1623Q channel by Kambouris et al. [111] have increased the propensity of R1623Q channels to inactivate without a significant change; n.d., not done.

The E1784K substitution is located in the highly conserved acidic proximal portion of the Na⁺ channel C-terminal domain, immediately following the DIVS6 segment. It is also associated with a small persistent inward Na⁺ current in the Xenopus oocyte [75] and tsA201 [105] expression systems. Steady-state inactivation of this mutant was shifted to more negative potentials [75,105]. In oocytes, this negative shift was exaggerated by co-expression with the β1-subunit [75]. In tsA201 cells a faster recovery from inactivation was also demonstrated [105]. The effect of this substitution is probably mediated through an allosteric mechanism, which disrupts critical conformational changes involved in channel gating.

The R1623Q mutation [41,109] in the DIVS4 segment is associated with a small persistent inward current, and a markedly slowed rate of macroscopic current decay caused by a decreased rate of transition from the open state to the inactivated state. The inactivation defect of this mutant channel may be attributed to uncoupling of inactivation activation coupling (Section 2.1.2).

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The DII–DIII linker S941N mutation was described recently in a case of LQT3 association (Section 2.1.2). Hence, a possible molecular mechanism markedly slowed rate of macroscopic current decay caused by a decreased rate of transition from the open state to the inactivated state. The inactivation defect of this mutant channel may be attributed to uncoupling of inactivation activation coupling (Section 2.1.2).

Pharmacological studies on lidocaine blockade of the R1623Q channel by Kambouris et al. [111] have increased our insight into the mechanisms of block of normal and LQT3-associated sodium channels. They demonstrated that rather than blocking the open channel, lidocaine augments the propensity of R1623Q channels to inactivate without a prior opening, that is, they transit more often from the closed state directly to the inactivated state (‘closed-state inactivation’). Kambouris et al. [111] propose that the effect of lidocaine on other LQT3-associated channels like

**Table 1**

<table>
<thead>
<tr>
<th>Mutation</th>
<th>Domain</th>
<th>Shift in V&lt;sub&gt;1/2&lt;/sub&gt; of activation</th>
<th>Shift in V&lt;sub&gt;1/2&lt;/sub&gt; of inactivation</th>
<th>Recovery from inactivation</th>
<th>Time-course of inactivation</th>
<th>Persistent I&lt;sub&gt;Na&lt;/sub&gt;</th>
<th>Single channel characteristics</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>S941N</td>
<td>DII–DIII linker</td>
<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
<td>0.25–0.4% (1.8–2.5× larger than WT)</td>
<td>Increase in dispersive reopenings</td>
<td>[101]</td>
</tr>
<tr>
<td>N1325S</td>
<td>DIII loop S4–S5</td>
<td>−6.4 mV&lt;sup&gt;b&lt;/sup&gt;</td>
<td>n.s.&lt;sup&gt;1&lt;/sup&gt;</td>
<td>n.s.&lt;sup&gt;1&lt;/sup&gt;</td>
<td>Slower&lt;sup&gt;b&lt;/sup&gt;</td>
<td>n.s.&lt;sup&gt;1&lt;/sup&gt;</td>
<td>1.25×τ&lt;sub&gt;slow&lt;/sub&gt;</td>
<td>Increase in dispersive reopenings and V&lt;sub&gt;dp&lt;/sub&gt; bursts&lt;sup&gt;2&lt;/sup&gt;</td>
</tr>
<tr>
<td>ΔKPQ&lt;sup&gt;1505–1507&lt;/sup&gt;</td>
<td>DIII–DIV linker</td>
<td>n.d.</td>
<td>−5.8 mV</td>
<td>n.d.</td>
<td>Faster&lt;sup&gt;2&lt;/sup&gt;</td>
<td>τ&lt;sub&gt;τ&lt;sub&gt;slow&lt;/sub&gt; 1.50×&lt;/sub&gt;</td>
<td>5–2×&lt;sup&gt;2&lt;/sup&gt;</td>
<td>Increase in dispersive reopenings and V&lt;sub&gt;dp&lt;/sub&gt; bursts&lt;sup&gt;2&lt;/sup&gt;</td>
</tr>
<tr>
<td>R1623Q</td>
<td>DIVS4</td>
<td>n.s.</td>
<td>n.s.</td>
<td>n.s.</td>
<td>Slower&lt;sup&gt;2&lt;/sup&gt;</td>
<td>τ&lt;sub&gt;τ&lt;sub&gt;slow&lt;/sub&gt; 3.3×&lt;/sub&gt;</td>
<td>0.51%; Increase dispersive reopenings</td>
<td>[41,109]</td>
</tr>
<tr>
<td>R1644H</td>
<td>DIVS4</td>
<td>n.s.&lt;sup&gt;1&lt;/sup&gt;</td>
<td>n.s.&lt;sup&gt;1&lt;/sup&gt;</td>
<td>Faster&lt;sup&gt;2&lt;/sup&gt;</td>
<td>n.s.&lt;sup&gt;1&lt;/sup&gt;</td>
<td>τ&lt;sub&gt;τ&lt;sub&gt;slow&lt;/sub&gt; 3.3×&lt;/sub&gt;</td>
<td>0.85%; Increase dispersive reopenings</td>
<td>[32,108]&lt;sup&gt;3&lt;/sup&gt;</td>
</tr>
<tr>
<td>E1784K</td>
<td>C-terminal</td>
<td>n.d.</td>
<td>−5.3 mV</td>
<td>n.d.</td>
<td>Slower&lt;sup&gt;2&lt;/sup&gt;</td>
<td>1.8×τ&lt;sub&gt;slow&lt;/sub&gt;</td>
<td>2×&lt;sup&gt;2&lt;/sup&gt;</td>
<td>Increase dispersive reopenings</td>
</tr>
<tr>
<td>+β1</td>
<td>n.d.</td>
<td>−12.1 mV</td>
<td>Faster&lt;sup&gt;2&lt;/sup&gt;</td>
<td>2×&lt;sup&gt;2&lt;/sup&gt;</td>
<td>n.s.&lt;sup&gt;1&lt;/sup&gt;</td>
<td>1.9×τ&lt;sub&gt;τ&lt;/sub&gt;</td>
<td>2±4%; Increase dispersive reopenings</td>
<td>[75]</td>
</tr>
<tr>
<td>D1790G</td>
<td>C-terminal</td>
<td>n.s.&lt;sup&gt;1&lt;/sup&gt;</td>
<td>n.s.&lt;sup&gt;1&lt;/sup&gt;</td>
<td>n.d.</td>
<td>Faster&lt;sup&gt;2&lt;/sup&gt;</td>
<td>1.3×τ&lt;sub&gt;fast&lt;/sub&gt;</td>
<td>n.s.&lt;sup&gt;1&lt;/sup&gt;</td>
<td>n.d.</td>
</tr>
</tbody>
</table>

<sup>a</sup> Changes and/or shifts are relative to the wild-type channel. Persistent I<sub>Na</sub> is given as a percentage of peak current for that channel. N.s., no statistically significant change; n.d., not done.

<sup>b</sup> Values found in mammalian cell line expression systems; all other data are from the Xenopus oocyte expression system.
Table 2
Electrophysiological characteristics of mutant cardiac sodium channels associated with Brugada syndrome

<table>
<thead>
<tr>
<th>Mutation</th>
<th>Domain</th>
<th>Shift in (V_{1/2}) of activation</th>
<th>Shift in (V_{1/2}) of inactivation</th>
<th>Recovery from inactivation</th>
<th>Time-course of inactivation</th>
<th>Persistent (I_{Na})</th>
<th>Single channel characteristics</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>R1512W</td>
<td>DIII–DIV linker</td>
<td>-5.1 mV</td>
<td>-3.8 mV</td>
<td>Slower</td>
<td>1.2× at -100 mV</td>
<td>n.d.</td>
<td>n.d</td>
<td>[160]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>n.s. (^b)</td>
<td>n.s. (^b)</td>
<td></td>
<td></td>
<td>n.d.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>T1620M</td>
<td>DIV S3–S4 loop</td>
<td>n.s.</td>
<td>+10 mV or +4.8 mV</td>
<td>Faster</td>
<td>n.d.</td>
<td>n.d.</td>
<td></td>
<td>[3,157, 158]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>n.s. (^b)</td>
<td></td>
<td></td>
<td></td>
<td>n.d.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>+β1</td>
<td></td>
<td>+10.5 mV</td>
<td></td>
<td>Faster</td>
<td>3× at -80 mV</td>
<td>n.s.</td>
<td></td>
<td>[157]</td>
</tr>
<tr>
<td>+β1(^b)</td>
<td></td>
<td>n.s. (^b)</td>
<td></td>
<td></td>
<td></td>
<td>n.s.</td>
<td></td>
<td>[158]</td>
</tr>
<tr>
<td>32–37°C</td>
<td></td>
<td>+10.7 mV (^b)</td>
<td>n.s. (^b)</td>
<td>Slower</td>
<td>32°C, -110 mV</td>
<td>Faster(^b)</td>
<td>n.d.</td>
<td>[158]</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>2× at 37°C</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1795InsD</td>
<td>C-terminal</td>
<td>+8.1 mV</td>
<td>-7.3 mV</td>
<td></td>
<td></td>
<td>n.d.</td>
<td></td>
<td>[120]</td>
</tr>
<tr>
<td>(Brugada+LQTS)</td>
<td></td>
<td></td>
<td></td>
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<td>n.s. (^b)</td>
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<td>A1924T</td>
<td>C-terminal</td>
<td>-9 mV</td>
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<td>n.d.</td>
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<td>[160]</td>
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</table>

\(^a\) Changes and/or shifts are relative to the wild-type channel. Persistent \(I_{Na}\) is given as a percentage of peak current for that channel. N.s., no statistically significant change; n.d., not done.

\(^b\) Values found in mammalian cell line expression systems; all other data are from the Xenopus oocyte expression system.

The sustained inward current caused by the S941N, ΔKPQ, N1325S, R1644H, R1623Q and E1784K mutations potentiates the small depolarizing Na\(^+\) current at the plateau phase of the cardiac action potential [112]. This provides a direct explanation for the delayed ventricular repolarization in LQTS patients harbouring these mutations. Moreover, the slower current decay of R1623Q channels (Table 1) may augment the effect of sustained current on the plateau potential prolongation, and the faster recovery from inactivation of ΔKPQ, R1644H, and E1784K channels (Table 1) may contribute to arrhythmogenesis.

However, one of the LQTS\(_s\) mutations characterised to date, the D1790G mutation, is not associated with a persistent inward current [76]. This mutation appears to alter α–β1 subunit interaction. Voltage-dependence of steady-state inactivation of monomeric D1790G channels was similar to that of monomeric wild-type channels. However, whereas in the case of the wild-type channel co-expression with the β1-subunit shifted the steady-state inactivation by 11 mV to more positive potentials, co-expression of D1790G mutant channels with the β1-subunit failed to produce such a shift. The mechanism by which this mutation prolongs repolarization is, however, unclear. It has been suggested that the mutation prolongs the action potential indirectly by alteration of the balance of net plateau currents secondary to a reduction in inward Na\(^+\) current during the action potential upstroke [113]. The slower upstroke and the less positive overshoot of the action potential during the very early stages of ventricular depolarization could cause an increase of Ca\(^2+\) entry via L-type Ca\(^2+\) channels. The alteration in Ca\(^2+\) transient that follows could subsequently affect calcium-sensitive processes thereby resulting in net increase of inward plateau current and correspondingly increase in action potential duration [113].

SCN5A transcripts have recently been demonstrated in limbic circuitry of rat brain [114]. It may thus be speculated that patients in whom prolonged QT interval presents in conjunction with epileptic seizures [115], may harbour mutations in SCN5A.

3.2. Clinical features of LQTS\(_s\) and pharmacological intervention

Whereas functional studies on almost all SCN5A mu-
tants associated with LQTS₃ have been published (Section 3.1), there is a remarkable paucity of clinical data on mutation carriers. Data from the LQTS registry group demonstrate the occurrence of a higher percentage of lethal events in LQTS₃ patients compared to LQTS₁ and LQTS₂ patients [116]. Events in general occur at a relatively later age in LQTS₁ patients [116]. In contrast to LQTS₁ and LQTS₂, QT-prolongation is equal in males and females and there is no age-difference [117]. Bradycardia could be part of the clinical picture, at least for some mutant channels. Patients carrying the ΔKPQ mutation have significant resting bradycardia and maximum exercise rate is attenuated as well ([118], A. Moss, personal communication). There is a trend towards lower heart rate in D1790G carriers and sinus arrest has been documented in three carriers of this mutation [76,119]. In the family described with the E1784K mutation, the proband’s father, who also carried the mutation, had sinus bradycardia (32 bpm) and occasional sinus pauses [75]. Decreased heart rate and occurrence of patients with asystolic episodes are also the case in carriers of the 1795insD SCN5A mutation described in a kindred with a combined LQTS₁-BS phenotype [120,121] (see Section 4.2). In the latter patients, but also in ΔKPQ and D1790G carriers, prolonged conduction parameters at different cardiac levels are reported [119–123].

LQTS₃ mutations are associated with a gain-of-function defect, and it is therefore immediately apparent that a gene-specific therapeutic approach targeted at blocking the persistent inward Na⁺ current responsible for action potential prolongation in this form of the disorder could become feasible. Data from experimental models mimicking LQTS₃ as well as the very limited clinical data available, suggest that Na⁺ channel blockers such as lidocaine and its analogue mexiletine could constitute a specific therapy in patients with defects associated with sustained inward Na⁺ current. In mammalian expression systems, these Class IB agents are able to preferentially block the late-opening ΔKPQ [124,125], R1644H [125] and N1325S [125] mutant Na⁺ channels at a low concentration [125] without greatly modifying peak Na⁺ current. This implies that pharmacological shortening of QT-interval in carriers of these mutations could be possible without compromising the spreading of excitation through the heart.

The prolonged guinea pig ventricular myocyte action potential in an in vitro (anthopleurin A) model mimicking LQTS₃ was shortened by mexiletine [126]. In experiments on the arterially perfused wedge [127] and in epi-, endo-, mid-myocardial and transmural strips [128] from canine left ventricle pretreated with ATX-II as a surrogate of LQTS₁, mexiletine was capable of abbreviating QT-interval, reduce transmural dispersion of repolarization and prevent torsades de pointes. This effect was secondary to a preferential effect of the drug to abbreviate the action potential duration of the M cell and to suppress the development of early afterdepolarizations in these cells.

In a study on six LQTS₃ patients, namely ΔKPQ and R1644H carriers, Schwartz et al. [129] demonstrated that five had a significant shortening of QT-interval in response to mexiletine. Rosero et al. [130] reported shortening of QT-interval in two ΔKPQ carriers by short-term intravenous lidocaine and long-term tocainide therapy. Also, the single patient with the R1623Q mutation responded favourably on lidocaine [41]. The specificity of lidocaine block for LQTS₃, however, is doubted since QT interval also shortens in response to lidocaine in LQTS₂ patients [131]. The specificity of Na⁺ channel blockers for LQTS₃ has also been questioned by experimental studies showing that mexiletine is also effective in suppressing torsades de pointes in LQTS₁ and LQTS₂ models, due to an effect of the drug to reduce transmural dispersion of repolarization and despite a relatively small effect of the drug to reduce QT interval [127,132].

However, Class IB Na⁺ channel blockade may not constitute a universal therapeutic approach to LQTS₃. As predicted on the basis of the absence of sustained inward Na⁺ current in D1790G mutant channels [76], lidocaine did not correct QT-prolongation in the two D1790G carriers tested [119]. However, the Class IC agent flecainide induced significant shortening of repolarization parameters among carriers of this mutation [119]. A preliminary report suggested that this therapeutic effect could be due to sensitivity of mutant channels to flecainide use-dependent block and slower recovery from block [133]. Although there are also indications that flecainide could also be effective in ΔKPQ carriers (A. Moss, personal communication), it is still unclear whether flecainide can be used for all LQTS₃ defects. Nevertheless, at this stage it appears that pharmacological intervention should be restricted to a research environment where correlation between type of mutation, electrophysiological characteristics of the mutated channel and outcome of channel blockade is possible.

Theoretically pacemaker therapy is also likely to be very effective in management of LQTS₃, because attenuation of QT-interval prolongation is seen at faster rates both in LQTS₁ patients [129] and in experimental models mimicking LQTS₃ [126,127]. Indeed, in the family with the 1795insD mutation, long-term pacemaker therapy proved very effective [121].

4. Brugada syndrome

The Brugada syndrome [134], for review see Refs. [135] and [136] is characterized by ventricular fibrillation and sudden cardiac death associated with the electrocardiographic pattern of ST-segment elevation in leads V₁–V₃ (unrelated to ischemia, electrolyte abnormalities, or structural heart disease). The ECG pattern is dynamic and varies presumably with varying risk of developing ventricular fibrillation [137]. Right bundle branch block, left axis deviation and increased HV interval, indicative of
Right bundle branch block morphology (due to early repolarization of the right ventricle, see further) is also often observed. This syndrome is associated with a high mortality rate [137] and is closely related to the sudden unexpected death syndrome (SUDS [139]) described in male Thai patients and responsible for an annual mortality rate of 1/2500 among young males in Thailand. A high prevalence of this syndrome is also present among the Japanese. It displays an autosomal dominant inheritance [3] with incomplete penetrance [140,141]. It affects predominantly men, with the first arrhythmic event occurring during the fourth decade of life but could also occur in children even during the first months of life [141].

Thus far, mutations in SCN5A [3,142] have been linked to the disorder, however, genetic heterogeneity is suggested by the description of a family in whom there was absence of linkage to the SCN5A locus on chromosome 3 [143] as well as absence of SCN5A mutation in a cohort of patients as determined by single-strand conformation polymorphism analysis methodology (unpublished data).

4.1. Proposed mechanism for the ECG pattern and genesis of arrhythmias

A mechanism based on transmural differences in action potential morphology has been proposed by Antzelevitch and co-workers [144,145] (Fig. 2). Unlike the endocardial action potential, the epicardial action potential possesses a ‘spike-and-dome’ morphology caused by a pronounced transient outward ($I_{to}$) current-mediated phase 1 [146,147], which predisposes these cells to ‘all-or-none’ repolarization. An increase in net outward current or a decrease of inward current at the end of phase 1 in epicardial cells may explain the ST-segment elevation observed in Brugada patients. Augmentation of outward current or decrease of inward current at the end of phase 1 would lead to loss of the epicardial action potential dome and selective abbreviation of the epicardial action potential giving rise to a voltage gradient across the myocardium generating the ST-segment elevation observed in the ECG. In this setting arrhythmias could arise from phase 2 re-entry secondary to the increased epicardial and transmural dispersion of repolarization. The prominence of the ECG changes in the right precordial leads could be explained by the fact that right ventricular epicardial cells display a larger $I_{to}$ (and hence more pronounced ‘spike-and-dome’) than left ventricular epicardial cells [148]. Moreover, it can be hypothesised that because of the thinness of the right ventricular wall, the relative contribution of epicardial action potentials to the surface ECG is more prominent in right than left precordial leads [135].

Experiments by Antzelevitch et al. [149] on the canine right ventricle (on tissue, on myocytes from the respective regions of the wall and on arterially perfused wedges of right ventricle) involving modulation of currents (augmentation of outward current or suppression of inward current) at the end of phase 1 of the action potential, have provided direct evidence that accentuation of epicardial—

![Fig. 2. Putative epicardial and endocardial action potential morphology, sodium current ($I_{Na}$) characteristics and surface ECG in normal subjects, patients with Brugada syndrome and patients with Long QT syndrome type 3. Note the abbreviated epicardial action potential and decreased $I_{Na}$ as proposed by Charles Antzelevitch as the basis of ST-segment elevation in Brugada syndrome. Note the prolonged epicardial and endocardial action potential, the altered $I_{Na}$ characteristics (slower inactivation, persistent inward current) and prolonged QT-interval in long QT syndrome type 3.](image-url)
endocardial action potential heterogeneity underlies the development of ST-segment elevation and the genesis of arrhythmias.

In man, Class I A Na⁺ channel blockers such as procainamide and ajmaline, can induce an elevation of the ST-segment in the early precordial leads or can increase pre-existing ST-segment elevations in patients with the Brugada syndrome [138,150] see [151]. Class IC Na⁺ channel blockers such as flecainide have also been shown to induce ST-segment elevation [152,153]. In fact, pharmacological suppression of $I_{Na}$ is often used to unmask the Brugada ECG in patients previously recognised but with transient normalization [154]. Sodium channel blockade has also been proposed to unmask the disorder in relatives of affected individuals to identify those at risk [154], although a study by Priori et al. [140] on the other hand, has demonstrated a lack of correlation between genetic status and the response to the flecainide test in asymptomatic gene carriers. Discussion has also been started concerning the management of patients in whom Class IC antiarrhythmic agents induce J-point elevation [151,153]. As yet no mutations have been identified in these latter patients.

There are also profound effects of autonomic stimulation and blockade on ST-segment elevation in Brugada patients [138]. Acetylcholine and β-blockers increase and isoproterenol mitigates ST-segment elevation.

4.2. SCN5A mutations in the Brugada syndrome

Since a decrease in Na⁺ channel density and conductance was considered to constitute a probable pathophysiological mechanism for the disorder, a candidate gene approach directed at SCN5A led to the identification of SCN5A mutations that co-segregated with the disorder in small families [3] and in sporadic cases with the disorder [142]. Two types of mutations have been described (Fig. 1, Table 2): mutations possibly leading to a decrease of functional Na⁺ channels at the sarcolemma, and missense mutations that give rise to Na⁺ channels with altered biophysical properties. The former category of mutations include an insertion of two nucleotides (AA) which disrupts the 5′ splice site of intron 7 [3], a deletion of a single nucleotide (A) at codon 1397 (frameshift mutation) leading to the creation of an in-frame stop codon and the generation of a truncated protein lacking part of DIII, DIV and the C-terminus [3], and a missense mutation (R1432G) residing in the P-loop of DIII [105]. Injection of SCN5A cRNA encoding the frameshift mutation into *Xenopus* oocytes of channels $\beta$1-subunit co-expression). On the other hand, Dumaine et al. [159], who investigated the biophysical analysis of R1432G mutant channel in tsA-201 cells [105] failed to express Na⁺ currents. In the case of the splice site mutation, it has not been demonstrated yet whether this mutation actually results in aberrant splicing or to what extent normal splicing is compromised.

A reduction in Na⁺ channel density is expected to reduce the action potential amplitude, which in turn leads to a negative shift in the voltage at which phase 1 repolarization begins. In experimental systems, flecainide has been shown to selectively shorten the epicardial action potential as a result of both this shift and the pronounced $I_{Na}$ in epicardial cells [155], and cause phase 2 re-excitation. Moreover, a decreased $I_{Na}$ could account for the conduction disturbances observed in a substantial subset of patients. In addition, it could be hypothesised that patients heterozygous for such ‘loss-of-function’ mutations have a 50% reduction in Na⁺ channels and a similar reduction in $I_{Na}$. However, this is very unlikely and compensatory mechanisms are anticipated to play a role since such a drastic reduction in Na⁺ current would have disastrous consequences on the action potential.

The other mutations associated with the disorder (Fig. 1, Table 2) are an L567Q substitution [141] and a T632M substitution [156] both located in the DI–DII linker, an R1512W substitution [142,105] located in the DIII–DIV inactivation gate, a T1620M substitution [3] located within the S3–S4 linker of DIV and an A1924T substitution [142] in the distal part of the cytoplasmic C-terminal domain.

Functional analysis in *Xenopus* oocytes of channels carrying the T1620M substitution [3,157,158] demonstrated a faster recovery from inactivation [3,157,158] and a positive shift in the inactivation curve [3,157,158]. Moreover, co-expression with the $\beta$1-subunit further shifted the mid-point of steady-state inactivation with respect to the wild-type channel [157] and accelerated further the recovery from inactivation [157,158]. The shift of the inactivation curve to more positive potentials (with normal activation kinetics) results in a larger window current and it has been argued that this could induce hyperexcitability and an arrhythmogenic substrate in BS patients [157]. These changes do not readily explain the electrocardiographic phenotype (see also below). In order to gain further insight into the mechanism whereby this mutant channel causes ST-segment elevation, two separate groups of investigators [158,159] have examined this channel in the mammalian cell line expression system. Baroudi et al. [158] demonstrated a slower recovery from inactivation (which is exacerbated by $\beta$1-subunit co-expression). On the other hand, Dumaine et al. [159], who investigated the characteristics of the mutant channel at a temperature (32°C) closer to the physiological range described faster decay and slower reactivation of the current of T1620M channels. The more rapid decay of $I_{Na}$ is expected to result in an outward shift of current flowing in phase 1, leading to abolishment of the action potential dome in epicardial cells. The temperature sensitivity of these changes reconciles with the sporadic cases where arrhythmogenic episodes occurred during febrile states. These investigators also found a positive shift of the steady-state activation curve that could account for slowed conduction in some patients.

Biophysical analysis of R1512W and A1924T mutant
channels in *Xenopus* oocytes [160] demonstrated a negative voltage shift of the steady-state activation and inactivation curves for R1512W channels and a negative voltage shift of the steady-state activation curve for A1924T. The mechanisms whereby these functional abnormalities give rise to the Brugada phenotype are yet unclear. The substantial negative voltage shift of the steady-state activation of mutant channels may lower the action potential threshold thereby increasing excitability. The increase in window current of these mutants (smaller in the case of the R1512W channel since voltage dependence of inactivation was also shifted to more negative potentials) may increase the depolarizing force early in the course of the action potential. Once again this does not explain the electrophysiologic characteristics of BS. As a highly speculative hypothesis one may forward that a sufficient increase in window current, as a result of the negative shift of the activation curve [160] of the mutant channels, may lead to depolarizing forces at resting membrane potential. Such depolarizations would lead to a larger fraction of inactivated channels, consequently decreasing the magnitude of the Na⁺ current during the action potential upstroke. Similar to mutations more directly leading to reduced inward \( I_{Na} \), these changes leave \( I_{to} \) unopposed, potentially leading to abbreviation of the epicardial action potential.

When analyzed in tsA-201 cells [105], voltage-dependence of steady-state activation and inactivation of the R1512W channel were not different from the wild-type channel. In this expression system however, the mutant channel displayed a slowing of inactivation and a slowing of recovery from inactivation expected to result in decreased channel density available for opening after each cardiac cycle giving rise to conduction abnormalities.

More experiments are needed to gain further insight into the exact mechanism responsible for the electrocardiographic features of this disorders and the genesis of arrhythmias. In addition, it is becoming increasingly clear that caution should be exerted in the interpretation of characteristics of ion channels expressed in heterologous expression systems and extrapolation of observations made in these systems. For example, channel kinetics are dependent on the expression system used [161] (also for example compare electrophysiological properties of the R1512W and T1620M channels expressed in oocytes to those obtained in mammalian cells, Table 2). The temperature at which channels are analyzed [159,162] is an important factor. Moreover, in mammalian cells, conditions that may vary could include variation in endogenous expression of the β1-subunit and its splice variants [163] as well as differences in ratio between exogenous/endogenous β1-subunit expression. Nevertheless, at times, different results have also been reported when experimental conditions seem comparable. For example, when comparing data reported by Dumaine et al. [159] and Baroudi et al. [158], both in tsA-201 cells, at ambient temperature and without exogenous coexpression of β1-subunit, findings for recovery from inactivation for the T1620M channel differ (Table 2).

Electrocardiographic features of the LQTS and the BS may occur together in the same patient. Recently, we have reported on a large 8-generation family characterized by a high incidence of nocturnal sudden death and electrocardiographic features of both the LQTS and the BS in the same patients [120]. A mutation in *SCN5A*, 1795insD, located in the acidic proximal portion of the cytoplasmic C-terminal domain, was identified in all electrocardiographically affected individuals and underlies the combined LQTS-BS phenotype. Heterologous expression in *Xenopus* oocytes of 1795insD mutant channels demonstrated a negative shift of the steady-state inactivation curve and a positive shift of the steady-state activation curve. The resulting reduction of open channel availability upon depolarization is expected to give rise to a reduced Na⁺ current during the action potential upstroke [120]. Such a reduction in inward Na⁺ current is in accordance with the Brugada features on the ECG and the slightly hampered conduction observed in some patients. Whereas in the oocyte expression system no sustained inward current was identified, a small sustained inward current compatible with the LQTS phenotype was found when this mutant channel was co-expressed with the β1-subunit in HEK 293 cells [164]. Detailed analysis of current kinetics in this system revealed that this mutation has opposite effects on the fast and slow components of inactivation. Fast inactivation is disrupted, causing the sustained inward current throughout the action potential plateau and prolonging repolarization at slow heart rates, whereas slow inactivation is enhanced, delaying recovery of channel availability between stimuli and reducing the Na⁺ current at rapid rates. This would lead to enhancement of the LQTS phenotype at slow heart rates and enhancement of the Brugada phenotype at higher rates. These findings were corroborated by ECG-recordings from a patient carrying the 1795insD mutation during rest and with exercise. QT-interval was prolonged and ST-segment elevation was moderate at rest. With exercise there was an increase of ST-elevation which closely correlated with increase in heart rate, and QT-interval was shortened [164].

### 4.3. Patient management

As far as therapy of BS patients is concerned the implantable cardioverter-debrillator (ICD) has been suggested as the only effective treatment [136]. Because of a similarly bad prognosis, ICD treatment is also advocated for asymptomatic patients with the typical ECG features [136]. On the other hand, long-term follow-up of isolated patients questions this aggressive approach [165,166]. EP-guided pharmacological treatment with Class IA drugs (quinidine in particular) seems to be safe and effective in previously resuscitated Brugada patients.
[166]. Indeed, experimental data support the concept that quinidine, by virtue of its $I_{to}$ blocking effect, might be effective in BS patients [167]. However, in view of limited patient numbers definite conclusions on the most appropriate treatment modality cannot be derived as yet. This holds in particular for the asymptomatic patients and for the asymptomatic carriers of disease-related mutations with a normal ECG.

5. Isolated cardiac conduction defects

We have recently demonstrated that cardiac conduction defects form a third cardiac disorder associated with mutations in SCN5A [4]. This also makes SCN5A the first gene identified whose mutation causes an isolated conduction defect.

SCN5A was found to be mutated in two families with different forms of the disease; a progressive form, also called Lenègre-Lev disease [168,169] or progressive cardiac conduction disorder (PCCD), and a non-progressive form. The family with the progressive form of the disorder was characterized by progressive hampered cardiac conduction through the His-Purkinje system with right or left bundle branch block and widening of QRS complexes, leading to complete atrioventricular block and causing syncope and sudden death. Linkage was demonstrated to the SCN5A locus and an SCN5A splice site mutation (IVS22+2 T→C) predicted to result in in-frame skipping of exon 22 and a product lacking the voltage-sensitive S4 segment of DIV was identified. A locus (HBN1) on chromosome 19q13.3 is also linked to the Lenègre-Lev disease [170,171]. The gene at this locus however has not yet been identified.

In the family with the non-progressive form of conduction disorder, the proband and a brother presented after birth with an asymptomatic first-degree atrioventricular block associated with right bundle branch block. The mutation in this family was a deletion of a single nucleotide (5280delG) that results in a frame-shift predicting disruption of S6 of DIV and truncation of the entire C-terminal domain of the protein.

Both mutations are expected to be associated with decreased $I_{to}$ either through a reduction of Na$^+$ channels at the sarcolemma or channels associated with drastically diminished conductance. However, delineation of the exact pathogenic mechanism for SCN5A mutations in this disorder awaits expression studies of these mutant channels.

6. Synopsis

Mutations in the cardiac Na$^+$ channel have been hitherto associated with three different forms of cardiac electrical disease, with sudden cardiac death as a common denominator. The three phenotypes (LQTS$_3$, BS, PCCD) share clinical features, including conduction defect (PCCD by definition, LQTS$_3$ and BS), nocturnal sudden cardiac death (BS, LQTS$_3$) that in part seems related to bradyarrhythmias, and repolarization disorder (BS, LQTS$_3$). This suggests that SCN5A-related disorders are more closely related than heretofore appreciated. Careful clinical evaluation may unmask more overlap in phenotype/s associated with the different mutations and all mutations might find their appropriate place in this scheme.

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