Catecholaminergic Polymorphic Ventricular Tachycardia

Running title: Leenhardt et al.; Catecholaminergic Ventricular Tachycardia

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Introduction:

Sudden death due to cardiac arrhythmias in the young is a devastating event and remains underdiagnosed. The primary electrical disorders responsible for polymorphic ventricular tachycardia (VT) or ventricular fibrillation (VF) are long QT syndrome (LQTS), Brugada syndrome, the short-coupled variant of torsades de pointes, short QT syndrome and catecholaminergic polymorphic VT (CPVT). CPVT is a rare arrhythmogenic disorder characterized by adrenergic-induced bidirectional and polymorphic ventricular tachycardia (VT). The prevalence of the disease is estimated to be 1:10,000 in Europe. The first case was reported in 1975 followed by our first series of patients. Key features include polymorphic VT reproducibly induced during exercise test, isoproterenol infusion, or emotion and exercise. CPVT occurs in children and adolescents and causes syncope and sudden cardiac death at a young age, in absence of structural heart disease. The resting electrocardiogram (ECG) including the QTc interval, is normal. The mortality of CPVT is extremely high reaching 31% by the age of 30 years when untreated. The estimated 4- and 8-year cardiac event rates were 33% and 58%, respectively, in our series of patients without beta-blockers. There is a clear correlation between the age of the first syncope and the severity of the disease, with a worst prognosis in case of early occurrence. Beta-blockers without sympathomimetic activity are clinically effective in reducing syncope. However, arrhythmic event rate on beta-blocker therapy remains significant, suggesting the need of alternate pharmacological and non-pharmacological therapies that will be discussed.

With the advancements of the molecular genetics and the identification of mutations in the genes encoding the cardiac ryanodine receptor and in cardiac calsequestrin 2 in patients with CPVT, the central role of the intracellular calcium dysregulation in myocardial cells is
progressively better understood through expression studies and murine models. Therapies targeting this dysregulation are actually developed.

**Genetic background**

The family history of syncope or sudden death in the initial reports was suggestive of a genetic origin. This was established with the description of two large Finnish families with typical presentation of CPVT inherited on an autosomal dominant mode and the identification of the first locus on chromosome 1q42-43 in 1999. Priori et al and Laitinen et al identified the first mutations in the cardiac ryanodine receptor gene (RYR2), in families suffering of this type of CPVT, now known as CPVT1. A recessive form has been described by Lahat et al in families belonging to a Bedouin tribe, and mapped to chromosome 1p13-21. They identified a homozygous missense mutation in the cardiac calsequestrin gene (CASQ2) as the cause of this recessive form, now known as CPVT2. We then described patients with homozygous nonsense CASQ2 mutation suggesting that complete absence of CASQ2 in humans is not lethal and does not induce apparently any structural abnormality. RYR2 mutations are frequent while CASQ2 mutations are rare; altogether mutations are only found in 50 to 60% of the CPVT patients, which suggests that other genes are involved. Recently, a new locus on chromosome 7p14-p22 was reported in an inbred Arab family with an early onset lethal form of recessive CPVT.

**Ryanodine receptor**

The cardiac ryanodine receptors (RyR2) are calcium (Ca\(^{2+}\)) release channels present in the sarcoplasmic reticulum (SR), an intracellular vesicular network playing a major role in the regulation of Ca\(^{2+}\) homeostasis in the heart. The mechanism of their activation is called calcium induced calcium release (CICR) since it requires that Ca\(^{2+}\) provided by the activated L-type Ca\(^{2+}\) channel (Cav1.2). Calcium binds to RyR2 and triggers opening of a high-conductance channel...
allowing rapid Ca\(^{2+}\) efflux from the SR. The consecutive high cytoplasmic Ca\(^{2+}\) induces myocardial contraction, then Ca\(^{2+}\) is reuptaked in SR where it is stored at high concentration. This cycle is finely regulated and its dysfunction is associated with cardiac diseases such as CPVT, sudden death, and heart failure\(^{12}\).

RyR2 is a homotetramer; each monomer contains an enormous cytoplasmic domain that serves as a scaffold for regulatory subunits and enzymes that modulate the function of the channel\(^{13}\), and a channel domain located at the carboxy terminus. Each monomer has at least 6 transmembrane segments forming the pore region of the channel\(^{14,15}\).

Many proteins associated directly or indirectly with the N-terminal cytoplasmic domain of RyR2 including the 12.6 kDa FK506-binding protein (calstabin-2 or FKBP12.6)\(^{16}\), protein kinase A (PKA)\(^{17}\), calcium/calmodulin-dependent kinase II (CaMKII)\(^{18}\), phosphodiesterase 4D3 (PDE4D3)\(^{19}\), calmodulin (CaM)\(^{20}\), protein phosphatases 1 and 2A (PP1 and PP2A)\(^{17}\), and sorcin\(^{21}\). Calsequestrin, junctin, and triadin are linked to the C-terminus of RyR2\(^{22}\).

RyR2 shares close to 70% with two other mammalian RyR isoforms\(^{15}\): RyR1 and RyR3. RyR1 is predominantly found in skeletal muscle where it is activated directly by the L-type Ca\(^{2+}\) channel (Cav1.1) to release SR Ca\(^{2+}\) stores during skeletal muscle contraction. Mutations in \(RYR1\) gene cause various muscle disorders such as malignant hyperthermia or central core diseases\(^{23}\).

**Ryr2 mutations**

The 4,967 amino acid RyR2 channel is encoded by one of the largest genes in human genome containing 105 exons. To date, more than 150 mutations have been reported, most of them for CVT1 and unexplained or exercise-induced sudden death (review in\(^{24,25-29}\)). A few mutations have been identified in patients described as presenting with type 2 arrhythmogenic right
ventricular cardiomyopathy (ARVC2)\textsuperscript{30} sudden infant death syndrom\textsuperscript{31} or even associated with QT prolongation (Figure 1). Most of RYR2 mutations are missense mutations, occurring in three hot spots regions, the N-terminal region, the central region where is localized the calstabin-2 binding domain, and the C-terminal domain, including the channel region. These 3 regions are well conserved among RYR gene family, and involved in regulation of RyR channels. Similar mutation clustering is observed for RYR1-disease associated mutations\textsuperscript{23}.

A recent screening of the 105 exons in a large cohort of CPVT patients has confirmed these hot spots\textsuperscript{24}. However, mutations are reported out of these hot spots, especially between domains I and II\textsuperscript{24,29,32}. If it is logical to screen first the most frequently mutated exons, and then all the exons with known mutations, but new mutation may occur in other exons, and thus necessitate performing a complete gene analysis. In addition, some large deletions have been reported\textsuperscript{24,26,33} and large genomic rearrangements may be much more frequent that thought since they are not explored with the techniques used in the diagnostic laboratories. When a mutation is detected, siblings and parents have to be screened for the mutation, even if they are asymptomatic. For genetic counselling it is important to note, that \textit{de novo} mutations are frequent (at least 20\% to 50\% in sporadic cases). Moreover, even if it is a rare event, germinal and somatic mosaicism may occur in an asymptomatic parent as reported in two studies\textsuperscript{24,28}. Somatic mosaicism is not investigated in most of the clinical diagnostic laboratories and requires additional tissues samples to analyze parental urinary cells, hair roots or buccal epithelium DNA. Germinal mosaicism is only investigated when the same mutation is identified in two, siblings and not identified in the parental blood DNA. It is cautious and easier, when a \textit{de novo} mutation is detected, to screen the siblings for the sporadic proband mutation, even if they are asymptomatic.
A high variability of the phenotypic expression among subjects of the same family or unrelated families was demonstrated and estimates of the penetrance range from 25% to 100% \textsuperscript{32,\hspace{0.5em}34-36}. Noteworthy, there are asymptomatic RYR2 mutation carriers with normal exercise stress tests. Some of them can further present with exercise-induced arrhythmia during subsequent stress test\textsuperscript{32}, but more importantly may die suddenly as the first manifestation of the disease\textsuperscript{34,\hspace{0.5em}35}. No genotype-phenotype correlations have been established so far, even if there are mutations identified in large pedigrees supporting a lower penetrance\textsuperscript{32,\hspace{0.5em}34}. In a few patients, two mutations have been reported\textsuperscript{32,\hspace{0.5em}37} and the role of associated polymorphisms, either frequent such as Q2958R, G1886S and G1885E, or more rare such as A1136V is not known\textsuperscript{29}. They may affect Ca\textsuperscript{2+} regulation as suggested by \textit{in vitro} studies and increase the risk of sudden death in some patients\textsuperscript{38}.

**CASQ2**

CASQ2 is the major intra-SR Ca\textsuperscript{2+}-binding protein and it is localized at the junctional face membrane in the SR\textsuperscript{39}. It is a highly acidic protein with numerous charged residues, and binds Ca\textsuperscript{2+} ions with low affinity\textsuperscript{40}. CASQ2 exists in the SR as a dynamic structure formed of monomers, dimers or multimers, depending of the Ca\textsuperscript{2+} concentration. While the multimeric forms of CASQ2, formed at high Ca\textsuperscript{2+} levels \textit{in vitro}, function as a Ca\textsuperscript{2+} buffer, the monomers appear to modulate SR Ca\textsuperscript{2+} release by influencing the open probability of the RyR2 channel, via interactions with triadin and/or junctin\textsuperscript{41}. Triadin and junctin are structurally homologous proteins with a single transmembrane domain and a long highly positively charged C-terminal domain extending in the lumen of the SR and involved in protein-protein interaction, especially with CASQ2. The SR luminal Ca\textsuperscript{2+}-dependent control of RyR2 activity by CASQ2 normally limits RyR2 open probability and contributes to RyR2 deactivation and to the development of a
temporary refractory state that occurs after each Ca\(^{2+}\) release. Studies on cells or myocytes after overexpression of mutant proteins and on various models of genetically modified mice deficient in Casq2 or triadin have repeatedly shown that CASQ2 is an important regulator of SR Ca\(^{2+}\) release \(^{42}\). CPVT mutations reduce the extent and shortening the duration of Ca\(^{2+}\) signalling refractoriness, increase RyR2 open probability, thereby promote SR Ca\(^{2+}\) release, and thus contribute to the genesis of the arrhythmias \(^{43,44}\).

This implies that CASQ2 truncating mutations, or missense mutations affecting either its polymerisation, or its interactions with RyR2, triadin, and possibly other proteins could deregulate the calcium release machinery and induce lethal arrhythmia under stress conditions.

**CASQ2 mutations**

The 399 amino acid CASQ2 protein is encoded by a gene containing 11 exons. Twenty-one distinct CASQ2 mutations have been reported, either homozygous or compound heterozygous mutations transmitted under a recessive mode of inheritance (Figure 2). Half of them are missense mutations localized in different exons \(^9,45-50\). The others lead to truncated proteins by various mechanisms, nonsense codon, small deletion and abnormal splicing leading to premature stop codon \(^10,35,46,51,52\). Interestingly, a synonymous c.381C>T variation in exon 3, recently identified in a CPVT2 family, was shown to induce abnormal splicing and a premature stop codon using a splicing minigene assay \(^52\).

The phenotype is similar between the patients with two CASQ2 mutations and the patients with a RYR2 mutation. Most of the carriers of a single CASQ2 mutation are healthy. Nevertheless, several clinical investigations suggested that a single CASQ2 mutation could represent a potential susceptibility for ventricular arrhythmias in some subjects \(^10,28,45\). The origin of the variability among subjects of a same family is still unknown. Two non synonymous
polymorphisms, Thr76Ala and Val76Met, have been described in CASQ2, both in Caucasian and Asian populations, but their possible mild functional effect has not been studied to our knowledge. Variants in the multiple proteins of the calcium release complex may also contribute to this individual susceptibility.

Pathophysiological background

CPVT1 and CPVT2 mutations result in inappropriate calcium leakage from the SR, leading to cytosolic calcium overload generating delayed afterdepolarizations (DADs), triggered activity and ventricular arrhythmias, in particular under adrenergic conditions. RyR2 present three sites of phosphorylation on serines, S2808, S2814 and S2030. During stimulation with isoproterenol, S2808 is phosphorylated by protein kinase A (PKA) and S2814 phosphorylated by CaMKII. The group of Marx and colleagues have shown that, during adrenergic stimulation, PKA increases the open probability of RyR2 by the phosphorylation of serine 2808 and the subsequent dissociation of calstabin-2. Whereas a general consensus exists that adrenergic stimulation increases spontaneous Ca\(^{2+}\) release, and that this leak is amplified in the presence of CPVT1 or CPVT2 mutations, the role of RyR2 phosphorylation and calstabin dissociation remains controversial.

There is increasing amount of data, provided by a mouse model of CPVT, showing a Purkinje origin of the ventricular premature beats. In this setting, it has been shown that DADs caused by calcium overload are of more common occurrence in Purkinje cells than in ventricular myocytes, both at baseline and after beta-adrenergic stimulation. The Purkinje cells are probably critical contributors to arrhythmic triggers in animal models and humans with CPVT.

Clinical presentation

CVPT was first described in 1975 by Reid and then in 1978 by Coumel who reported a series of
children without cardiac disease presenting with reproducible exercise-induced ventricular polymorphic arrhythmia\textsuperscript{1,2}. In 1995, our group studied a cohort of 21 patients with a seven-year follow-up and further refined the description of this entity in 2009 with a cohort of 101 patients\textsuperscript{3}. CVPT is extremely uncommon before the age of 2 years, Some RyR2 mutations have been identified post mortem in cases of Sudden Infant Death Syndrome\textsuperscript{31}. However, as CPVT-related documented arrhythmias at this very young age has never been reported, these genetic findings may not be causal of this disease. The first episode of syncope usually occurs during the first or second decade of life. The symptoms are always triggered by exercise or emotional stress. Typically, the clinical presentation is a syncope often associated with seizure induced by exercise or emotional stress. Often, epilepsy is diagnosed and children are inappropriately treated with long-term anti-epileptic therapy. A mean delay in diagnosis of 2 years or more is usually reported in patients with syncope initially attributed to vasovagal or neurological causes. A family history of exercise-related syncope, seizure or sudden-death is reported in 30% of the patients. Family screening is mandatory, as CPVT is an autosomal dominant disease. Asymptomatic carriers of a \textit{RYR2} mutation are often detected during screening of the family members of an index patient.

**Diagnosis**

A history of exercise- or emotional stress-induced syncope with polymorphic ventricular arrhythmia in a child is highly suggestive of CVPT, although some LQTS1 patients can have a similar presentation. The resting ECG is normal and the QT interval duration is normal but can be borderline in some cases. A lower than normal heart rate has been reported particularly in boys with \textit{RYR2} mutations\textsuperscript{3,35}. The heart is structurally normal. The arrhythmia is reproducibly induced during exercise test as well as during isoproterenol infusion. Holter monitoring or exercise test can document CVPT by showing the ventricular arrhythmia progressively appearing
after a heart rate threshold (around 120-130 bpm). Polymorphic VT is usually not inducible by programmed ventricular stimulation. Implantable loop recorders can be useful to record CVPT in children with adrenergically triggered unexplained syncope. Molecular analysis has shown that there is a small group of CPVT patients (mutation carriers) with an apparently normal phenotype, even after exercise tests. Worryingly, some of these phenotypically normal CVPT patients do experience syncope and sudden death implying that an asymptomatic phenotype does not guarantee protection from polymorphic VT. Our recent report also demonstrated that cardiac and lethal (or near lethal) event rates were similar between 50 probands and 51 affected family members, suggesting the fact that, in the family of newly diagnosed CPVT proband, identification of the affected relatives is mandatory. 

**Electrocardiographic key features in CVPT**

The resting ECG is usually normal and there is progressive ventricular ectopy as the heart rate increases during exercise or isoproterenol infusion. The frequency and complexity increases as the heart rate increases, first monomorphic ventricular premature beats (VPBs) followed by bidirectional VT (Figure 3). VPBs have usually a right bundle branch block pattern with alternating right and left axis deviation, suggesting a left ventricular origin. If the exercise is continued, salvos of polymorphic VT may appear, become more sustained and rapid, leading to syncope. Usually, the arrhythmia is self-terminating but in some cases it can degenerate into VF and sudden death (Figure 4). The arrhythmia disappears with the discontinuation of the exercise or after cessation of the isoproterenol infusion. The reverse heart rate-dependent sequence is usually observed during recovery. Some individuals expressing bidirectional VT during exercise may not have CPVT. Instead, clinical consideration of either Andersen-Tawil syndrome or LQTS and appropriate genetic testing may be warranted for RyR2 mutation negative individuals.
considered as CPVT patients, particularly females. Careful inspection of TU-wave morphology may assist in distinguishing between CPVT and Andersen-Tawil syndrome in a patient exhibiting exercise-induced bidirectional VT. Atrial arrhythmias, including atrial fibrillation, are not uncommon during exercise test, and have been described in some adult patients.

**Current management**

- **Beta-blockers**

The first-line therapeutic option for patients with CPVT is beta-blockers without sympathomimetic activity, in accordance with the arrhythmia’s catecholaminergic mechanism, combined with exercise restriction. Nadolol, being a long-acting drug is preferred for prophylactic therapy and has been found to be effective clinically. In our experience, the dosage used to provide adequate prevention of CVPT and syncope is usually high (1.8 mg/kg). We have reported in 2009 the long-term follow-up results of 101 CPVT patients with an estimated 8-year cardiac event rate of 27% even in those taking beta-blockers.

Numerous studies have reported the heterogeneous proportion of near-fatal and fatal arrhythmic events in CPVT patients. The apparent discrepancy in efficacy of beta-blocker treatment between the various studies probably reflects differences in genetic background, in beta-blocker dosages or a poor drug compliance. This discrepancy in beta-blocker efficacy may also be due to the presence of polymorphisms influencing their metabolism. Larger groups of CPVT probands are needed to address the issue of beta-blocker efficacy in CPVT.

Our study of 101 CPVT patients, showed that, a younger age at the time of the diagnosis and the absence of beta-blockers were independent predictors for cardiac events. A history of aborted cardiac arrest before the diagnosis and absence of beta-blockers were independent
predictors for life-threatening events. It’s worth noting that a history of syncope before the
diagnosis was not associated with higher cardiac or life-threatening event rates. In the subgroup
of 81 patients receiving beta-blockers, beta-blockers other than nadolol as well as a younger age
at the diagnosis were independent predictors for cardiac events.35

Meanwhile, the maximal well-tolerated dosages of beta-blocker should be prescribed and
Holter recordings and exercise tests should be repeated periodically to assure that the degree of
sinus tachycardia that precedes onset of arrhythmias is never reached. Moreover, once the
diagnosis is established it is crucial to make the patients aware of the necessity of the faultless
compliance to the beta-blocker therapy given the number of non-compliance related sudden
cardiac deaths. It is strongly suggested that genetically positive family members should receive
beta-blockers even after a negative exercise test.35

Asymptomatic VPBs usually persist on Holter recordings with an unmodified threshold
of appearance. Complete suppression of asymptomatic VPBs seems not to be mandatory. We
have identified that the presence of couplets or more successive VPBs during exercise testing
were significantly associated with future arrhythmic events (sensitivity 0.62; specificity 0.67)
suggesting to intensify the treatment in these cases.35

Implantable Cardioverter Defibrillator

An ICD implantation is recommended in CPVT patients with syncope and/or documented
sustained VT despite beta-blocker therapy.62 It should also be discussed in patients with a
history of aborted cardiac arrest, or those with poor beta-blocker tolerance or compliance.
Nevertheless, ICD can potentially have pro-arrhythmic effects in CVPT patients, as stress caused
by appropriate or inappropriate discharges could prove disastrous by evoking a self-inducing
vicious circle.63,64
However, a combination therapy, involving both an ICD and an optimized dosage of beta-blocker, should safeguard against any such adverse effects and provide ultimate protection in non-responsive patients. Flecainide and verapamil have been challenged in a small number of patients with promising results. An ICD should be considered in patients who do not respond to a combination of beta-blockers and flecainide or verapamil or in whom a left cardiac sympathetic denervation has been ruled out. Nonetheless, insertion of an ICD is a technical challenge in pediatric patients and problems such as inappropriate shocks, the need for a life-time protection requiring multiple reinterventions should be addressed when the decision is taken.

- **Verapamil**

Verapamil has been shown to be beneficial in some CPVT patients by reducing the ventricular arrhythmia burden on top of beta-blocker therapy during a short-term follow-up period.\(^{65,66}\)

- **Flecainide**

Flecainide has been proved to have RyR2 blocking properties and to reduce significantly the ventricular arrhythmia burden in two highly symptomatic CPVT patients.\(^{67,68}\) The efficacy of flecainide has been retrospectively confirmed in a multicenter study including 33 CPVT patients.\(^{67}\) The rationale to combine beta-blockers to flecainide was the persistence of ventricular arrhythmias and/or symptoms, while on beta-blocker alone or combined with a calcium channel blocker. Twenty-two (76%) patients had either a partial (n=8) or complete (n=14) suppression of exercise-induced ventricular arrhythmias by flecainide. No patient experienced worsening of exercise-induced ventricular arrhythmias. During a median follow-up of 20 months (range: 12-40) no arrhythmic events occurred, except for one patient who experienced ICD shocks for polymorphic VT, which were associated with very low flecainide levels. One patient was already arrhythmia free for over 25 years. However the flecainide...
efficacy in the prevention of arrhythmic events remains to be demonstrated prospectively on a long-term basis.

- **Left cardiac sympathetic denervation**

Short series have been published reporting significant results of the left cardiac sympathetic denervation (LCSD) on cardiac events. The first publication reported the efficacy of LCSD in 3 young CPVT patients, with a very long follow-up in two (20 and 10 years) in whom ventricular arrhythmias were not controlled by beta-blocker therapy.\(^{69}\) The following series reported results of LCSD in patients with resistant and symptomatic ventricular arrhythmias despite optimal pharmacological therapy.\(^{70-74}\) Although the short-term results seem encouraging, more data with a long-term follow-up are needed. The LCSD is not available in many centers all over the world as it requires a very well-trained surgeon and dedicated techniques. Therefore, the place of LCSD in the therapeutic management of CPVT patients resistant to optimal pharmacological therapy is actually unclear.

**Management of CPVT patients: Next steps**

The class 1c antiarrhythmic agent propafenone was recently reported to have similar RyR2 blocking properties to flecainide.\(^{70}\) It has been shown to prevent exercise-induced CPVT in CASQ2 mutated mice and to prevent ICD shocks in a 22-year-old CPVT patient who had been refractory to maximal standard drug therapy and bilateral stellate ganglionectomy.\(^{70}\) Propafenone might be a therapeutic option in resistant cases.

Other compounds, such as dantrolene\(^{71}\) (acting by stabilizing the leaky RyR2 through the correction of the defective inter-domain interaction) or JTV519\(^{72,74}\), a RyR2 channel inhibitor (acting as a ryanodine receptor modulator by improving the rebinding of calstabin-2 to hyperphosphorylated RyR2 channels.), and KN93, an inhibitor of calcium/calmodulin-dependent
protein kinase II are able to prevent exercise- and epinephrine-induced ventricular arrhythmias in CPVT mouse models. A prospective study with one of these compounds is actually ongoing in CPVT patients. More data are needed to validate these potential new therapeutic options in CPVT patients.

**Conclusion**

Early diagnosis of CVPT is crucial considering the high risk of sudden death in untreated patients and the relative good response to beta-blockers in the majority of cases combined to exercise restriction. Family screening by clinical evaluation and genetic testing is mandatory to identify undiagnosed patients and asymptomatic carriers who are at risk of cardiac events and should be treated. The place of the ICD is questionable considering the young age of the patients, its possible proarrhythmic effects, and the other pharmacological alternative therapies that have recently been proposed. However long-term efficacy data are awaited. Some new compounds as the RyR2 channel inhibitors are being prospectively studied. The therapeutic strategy in CPVT patients will possibly been modified in the next years thanks to the results of the ongoing studies.

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**References:**


**Figure Legends:**

**Figure 1.** Schematic diagram of the RyR2 protein with the 148 reported mutations. The most C-terminal ~500 amino-acids compose the transmembrane domain, while the major part is cytoplasmic. The mutations are largely clustered in three regions of the sequence (grey boxes): NH2-terminus, central domain, and C-terminal domain or channel region are denoted. Mutations initially described as ARVD2 mutations, a phenotype which has not been confirmed by other teams, are denoted by an asterisk, mutations identified in children who died from SIDS are underlined, and the mutation reported in a long QT family is double underlined. Some mutations occur in the 3 regions of divergence among the 3 RYR homologs: 1310-1423, 1815-1903.
(E1837K), 4208-4489 (Q4282V, V4298M, A4307C, G4315E, E4431K), that are proposed to underscore the functional differences between RYR isoforms.

**Figure 2.** Schematic representation of the CASQ2 gene, localization of the human mutations, and functional domains of the calsequestrin 2. Upper part: genomic organization of CASQ2 gene (68.77 Kb) with its 11 exons (boxes). The sizes are given in bp for exons and in Kb for introns. Nonsense (1), splicing (6) and deletions (4) mutations leading to putative non functional truncated proteins are given. The loss the acidic residues at the C-terminal domain, responsible for Ca^{2+} binding induce inability to bind Ca^{2+}, disruption of the formation of head-to-tail dimers and polymerization of CASQ2. Lower part: the coding sequence is shown with the missense mutations in purple and two SNPs in blue. Some known functional sites of the corresponding protein are shown by colored double arrows. The overall structure of CASQ2 is composed of three identical thioredoxin-like domains (green). Every domain has a highly conserved hydrophobic core with acidic residues on the exterior generating highly electronegative potential surfaces. The acidic C-terminal domain (356-399) includes another 28 negatively charged residues (yellow). In addition, the signal peptide (black), the junctional face membrane interaction domain (pink), the ANKRD2/CARP binding sites (dark blue), and the conserved sites among the various CASQ isoforms (red) are shown. Numerous residues are involved in front-to-front dimer interface, tetramer and higher-order linear polymers with back-to-back interface), depending on the ionic environment. The residues are involved in the front-to-front (dimers) and back-to-back interfaces (tetramers and polymers). They are the most highly conserved residues in the entire sequence. R33Q and D307H mutations result in monomers that appear to be unable to form properly oriented dimers.

**Figure 3.** 12 lead ECG tracing during stress test showing the typical aspect of bidirectional VT characterized by 180° alternating QRS axis on a beat-to-beat basis with a right bundle branch block pattern suggesting a left ventricular origin.

**Figure 4.** Holter tracings showing in a CPVT patient, pleomorphic and polymorphic VT preceding the occurrence of a VF.
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In the Genetic background:

Triadine is as a new gene involved in an autosomal recessive form of CPVT Three new mutations in the triadin gene (TRDN), a protein that links RyR2 and CSQ2 were found in a cohort of 97 CPVT patients, which cosegregated with the disease on a recessive mode of transmission in two families. Two TRDN mutations, a 4 bp deletion and a nonsense mutation, resulted in premature stop codons; the third mutation was a p.T59R missense mutation. The mutations identified led to the absence of the protein [1].

In the Management of CPVT patients: Next steps

A carvedilol analogue was recently shown to prevent stress-induced ventricular tachyarrhythmias in RyR2 mutant mice [2]. It was more effective when combined with a selective beta-blocker metoprolol or bisoprolol. No human data have yet been published with such drug association.

Catheter ablation:

Catheter ablation of the bidirectional ventricular premature beats that trigger ventricular fibrillation may become an adjunctive therapy in patients with refractory CPVT [3] The published experience is actually very limited.

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