Arrhythmia/Electrophysiology

Genetic Variation in Titin in Arrhythmogenic Right Ventricular Cardiomyopathy–Overlap Syndromes

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Background—Arrhythmogenic right ventricular cardiomyopathy (ARVC) is an inherited genetic myocardial disease characterized by fibrofatty replacement of the myocardium and a predisposition to cardiac arrhythmias and sudden death. We evaluated the cardiomyopathy gene titin (TTN) as a candidate ARVC gene because of its proximity to an ARVC locus at position 2q32 and the connection of the titin protein to the transitional junction at intercalated disks.

Methods and Results—All 312 titin exons known to be expressed in human cardiac titin and the complete 3’ untranslated region were sequenced in 38 ARVC families. Eight unique TTN variants were detected in 7 families, including a prominent Thr2896Ile mutation that showed complete segregation with the ARVC phenotype in 1 large family. The Thr2896Ile mutation maps within a highly conserved immunoglobulin-like fold (Ig10 domain) located in the spring region of titin. Native gel electrophoresis, nuclear magnetic resonance, intrinsic fluorescence, and proteolysis assays of wild-type and mutant Ig10 domains revealed that the Thr2896Ile exchange reduces the structural stability and increases the propensity for degradation of the Ig10 domain. The phenotype of TTN variant carriers was characterized by a history of sudden death (5 of 7 families), progressive cardiac dysfunction causing death or heart transplantation (8 of 14 cases), frequent conduction disease (11 of 14), and incomplete penetrance (86%).

Conclusions—Our data provide evidence that titin mutations can cause ARVC, a finding that further expands the origin of the disease beyond desmosomal proteins. Structural impairment of the titin spring is a likely cause of ARVC and constitutes a novel mechanism underlying myocardial remodeling and sudden cardiac death. (Circulation. 2011;124:876-885.)

Key Words: arrhythmia ■ arrhythmogenic right ventricular dysplasia ■ cardiomyopathy ■ death, sudden ■ genetics

A rrrhythmogenic right ventricular cardiomyopathy (ARVC) is an inherited myocardial disease characterized by fibrofatty replacement of the myocardium and a predisposition to cardiac arrhythmias. The most common presenting symptoms are palpitations, syncope, and sudden death. Structural and functional alterations to the right and left ventricles can also occur, leading to the development of heart failure. Recognized as a significant cause of sudden death in young persons and athletes, ARVC is frequently diagnosed postmortem. It is a familial disease in up to 50% of cases, and the predominant mode of transmission is autosomal dominant, except in the case of Naxos disease, in which palmoplantar keratosis and unusual wooly hair are also present owing to recessive mutations in the JUP and PKP2 genes.1–4 Low penetrance, age-dependent expression, and challenges in confirming a clinical diagnosis premortem, along with genetic and allelic heterogeneity, have all conspired to complicate the identification of “genetic cases,” especially in small pedigrees or when only a single individual is known or suspected to manifest the phenotype. Currently, a genetic defect can be confirmed in ≈40% to 50% of cases.3–7

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Currently, 12 different ARVC loci are reported in the Online Mendelian Inheritance in Man. Five of these known genes (DSP, PKP2, DSG2, DSC2, and JUP) encode proteins integral to cell-cell junctions at the intercalated disk, thereby defining the intercalated disks as a major player in ARVC pathogenesis. The role of 3 other genes that have emerged as candidates has been less well established to date: the growth factor TGFβ3, the ion channel subunit RYR2, and the transmembrane protein 43 (TMEM43) identified in the Newfoundland founder population.8–10 Currently, of the 4 remaining...
loci that are genetically linked to ARVC, a locus at chromosomal position 2q31.1-q32.2 (Online Mendelian Inheritance in Man No. 602087) is of interest because this 3.4-Mb linkage region defined by the D2S152 and D2S389 microsatellite markers is close to the cardiomyopathy gene titin (TTN) that sits 0.4 Mb centromeric to the ARVC4 locus. Furthermore, there is a functional link between the desmosome and titin because the titin filament connects to the transitional junction at the intercalated disk, and animal models with altered intercalated disks show sarcomere and Z-disk changes. Thus, TTN is a candidate gene for ARVC.

Titin, the largest protein in mammals, is expressed in both cardiac and skeletal muscle. Several isoforms ranging in size from 2970 to 3900 kDa are produced from the single TTN gene, which is located on chromosome region 2q31 and comprises 363 exons. Titin filaments bridge the sarcomere along its longitudinal axis, overlapping end to end at the Z disk and M band at the amino and carboxy ends of titin, respectively, and forming a contiguous filament along the myofibril. The role of titin in cellular mechanics and signaling has recently been reviewed. Specifically, the spring-like properties of the sarcomere that underlie passive and restorative forces occurring after sarcomere lengthening or shortening, respectively, have been attributed to titin, and these characteristics promote the restoration of resting sarcomere length.

Elucidation of the genomic sequence of TTN has allowed genetic studies in skeletal and cardiac muscle diseases. Several mutations in the terminal exon of TTN result in titibial muscular dystrophy that does not involve any cardiac signs or symptoms. TTN missense mutations have also been reported in hypertrophic cardiomyopathy (Arg740Leu and Ser3799Tyr). Additional mutations have been found in dilated cardiomyopathy with 9 nonsynonymous and 1 frameshift mutation reported. Although large size and central position in sarcomeric architecture of titin have made it an attractive candidate for cardiomyopathy studies, the challenges of performing large-scale mutation screening studies of all 363 exons have limited the pace of genetic studies. Our group initiated a DNA resequencing project of TTN in a collection of 38 well-characterized ARVC families.

Methods

Patient Population

Probands from 38 ARVC families underwent TTN DNA resequencing provided by the University of Washington, Department of Genome Sciences, under US federal government contract N01-HV-48194 from the National Heart, Lung, and Blood Institute. Families were selected from the Familial Cardiomyopathy Registry, a multi-center, 3-decade-long ongoing project studying human hereditary cardiomyopathies; an additional 150 dilated cardiomyopathy probands were also sequenced and are reported separately. The diagnosis of ARVC was made on the basis of the 2010 update of the 1994 consensus criteria, and all available living subjects were evaluated by the investigators. Clinical data collected included medical history, family history, physical examination, ECG, echocardiogram, and whenever available, Holter monitoring, signal-averaged ECG, and histology from explanted heart or endomyocardial biopsy. Medical records from deceased subjects were reviewed when available. Informed consent was obtained from living subjects, and the local institutional review boards approved the protocol.

Table 1. Rare TTN Variants

<table>
<thead>
<tr>
<th>Family</th>
<th>Exon</th>
<th>Variant</th>
<th>Amino Acid Change</th>
<th>Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>TNRVD001</td>
<td>37</td>
<td>C29453T</td>
<td>Thr2896Ile</td>
<td>Spring</td>
</tr>
<tr>
<td>DNRVD006</td>
<td>97</td>
<td>A97341G</td>
<td>Tyr8031Cys</td>
<td>Spring</td>
</tr>
<tr>
<td>TNRVD303</td>
<td>108</td>
<td>C106734T</td>
<td>His8848Tyr</td>
<td>N2A</td>
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<tr>
<td>DNRVD008</td>
<td>298</td>
<td>T215598C</td>
<td>Ile16949Thr</td>
<td>A band</td>
</tr>
<tr>
<td>TNRVD027</td>
<td>305</td>
<td>G221380A</td>
<td>Ala18579Thr</td>
<td>A band</td>
</tr>
<tr>
<td>DRNVD023</td>
<td>312</td>
<td>G226177T</td>
<td>Ala19309Ser</td>
<td>A band</td>
</tr>
<tr>
<td>DNRVD011</td>
<td>357</td>
<td>C27284T</td>
<td>Pro30647Leu</td>
<td>M line</td>
</tr>
<tr>
<td>TNRVD027</td>
<td>362</td>
<td>T281801C</td>
<td>Met33291Thr</td>
<td>M line</td>
</tr>
</tbody>
</table>

DNA Sequence Analysis

Exons and peri-exonic regions of titin isoform N2A (NM 133378), along with additional exons unique to the principal cardiac isoform N2B (NM 003319), were amplified from genomic DNA with polymerase chain reaction. This covered 312 exons (311 expressed as titin protein) and the complete 3’ untranslated region. The polymerase chain reaction primers were Tm matched and designed from a masked reference sequence. Each primer pair was tiled with a universal M13 forward and reverse sequencing primer for subsequent sequencing. Once the regions were amplified, each polymerase chain reaction product was sequenced from the forward and reverse direction to provide double-stranded coverage. Sequencing was carried out with Sanger Big-Dye Terminator sequencing on capillary-based machines (AB 3730, Applied Biosystems, Foster City, CA). The sequencing traces were base called and assembled on the reference sequence, and PolyPhred was applied to identify sequence variants and to provide genotypes across the samples. Prior genetic studies excluded mutations in the ARVC-linked genes DSC2, DSG2, DSP, and PKP2 (data not shown).

TTN Mutation Analysis

Stringent criteria for the classification of a mutation as putatively disease causing included the following: variants uniquely identified in the patient cohort, predicted alteration of an amino acid sequence, evolutionary conservation of the particular residue altered, predicted tolerability of nonsynonymous changes, segregation among affected family members, and absence in 400 normal control chromosomes. Mutations detected were evaluated against known TTN single nucleotide polymorphisms in available databases, and mutations present in multiple other families in the entire cohort were considered common and unlikely to be pathogenic mutations. Nonsynonymous coding mutations were evaluated for putative functional effects with SIFT and PolyPhen analysis and scored as tolerant or intolerant; tolerant mutations were considered unlikely to be pathogenic. Testing of mutations in additional ARVC family members and in 400 chromosomes from ethnically similar healthy control subjects was done with pyrosequencing (PSQ 96MA, Biotage, Uppsala, Sweden).

Protein Analysis

The wild-type (WT) and mutated human Ig10 segments of the titin protein were produced as recombinant proteins in Escherichia coli. The WT and mutant proteins were separated on 7% native tris-glycine polyacrylamide gels supplemented with 17% sucrose and were used for nuclear magnetic resonance studies. Two-dimensional 1H-15N heteronuclear single quantum correlation spectra on 0.25-mmol/L samples were acquired at 22°C on a Bruker DRX600 equipped with cryogenic triple-resonance probes and processed with Topspin. Tryptophan residue fluorescence was studied with 25 μmol/L WT or mutant protein solution (50 mmol/L Tris/HCl, pH 7.5, 150 mmol/L NaCl) to which varying concentrations of guanosine chloride were added. Fluorescence was measured at 30°C on a Biotek Synergy 2 plate reader by use of a xenon flash lamp with 284/10-nm excitation and 340/30-nm emission filters. Assays were
done in triplicate. Proteolysis assays were carried out with trypsin (25:1 protein:trypsin ratio by weight) at room temperature, followed by gel electrophoresis and quantitative gel analysis.

**Results**

**Genetic Analysis**

Eight unique TTN nonsynonymous variants were identified in 7 unrelated ARVC probands (Table 1 and Figure 1); 1 proband (TSRVD027) who was a compound heterozygote had 2 TTN variants. The nonsynonymous variants were all considered intolerant by SIFT/PolyPhen analysis, were not previously reported in the database of single nucleotide polymorphism, were absent in 400 control chromosomes and an additional 300 chromosomes from patients with dilated cardiomyopathy, and were associated with the ARVC phenotype in our analysis of segregation within the families.

![Figure 1. Pedigrees of arrhythmogenic right ventricular cardiomyopathy (ARVC) families with rare TTN variants. Male and female individuals are indicated by squares and circles, respectively. Black shading indicates the individuals meeting full ARVC diagnostic criteria; gray shading, a suggestive cardiac history and/or history of sudden unexplained death (see also Table 2); and white squares/circles, unaffected individuals based on available family and medical history or, when TTN variant status is indicated, based on full clinical evaluation by the investigators. TTN rare variant status for tested individuals is indicated (+, present; −, absent); parentheses indicates inferred status. Probands are identified with an arrow.](http://circ.ahajournals.org/lookup/figure/878/888/1)
In family TSRVD001, a C39453T mutation (exon 37, Thr2896Ile; Figures 2 and 3) showed complete segregation of the mutation with the ARVC phenotype in 6 affected individuals (Figure 1). The Thr2896Ile mutation, absent in all other tested families and control subjects, was shared by 2 fifth-degree relatives with ARVC (III-1 and V-5), providing strong genetic evidence that this TTN mutation is linked to the ARVC phenotype. Multiple relatives in this family met diagnos-

Figure 2. Exon structure of the human titin gene with location of rare TTN variants in ARVC families (black font on white background) indicated by exon location and amino acid change (in parentheses). Shown are also previously identified variants associated with other cardiac diseases. Variants in exons 3, 14 (2 different variants), 18, 49 (4 different variants), 326, 335, and 358 have all been associated with dilated cardiomyopathy (blue); additional variants in exons 358 and 360 have been associated with fetal cardiomyopathy (black). For details, original citations, and variants associated with skeletal muscle myopathies, see the work by Bogomolovas et al. Red rectangle indicates immunoglobulin-like domain; white, fibronectin type 3 domain; blue, unique sequence; green, Z-repeat domain; yellow, PEVK domain; and black, titin kinase domain. Based on Genbank accession AJ27782 and the work of Bang et al.

Figure 3. A, Multiple sequence alignments with known Ig structures indicate that the mutated threonine in Ig10 is located in the short loop connecting the A’ and B β-strands. The hydrogen bond network between the A’ and G strands is important for Ig mechanical stability, and mutations in the A’-B loop have been shown to disrupt this stabilizing network. B, Schematic representation of human Ig10 using the homology modeling server ModWeb with Thr2896, shown with a ball-and-stick model. C, Histological section of the right ventricular wall of patient III-3 from family TSRVD001 (Thr2896Ile). C1, The myocardium is substituted by fatty tissue and a layer of subendocardial fibrous tissue (Azan; magnification ×2.5). C2, Fibrofatty infiltration with the presence of inflammatory cells (hematoxylin-eosin; magnification ×4.5.).
tic criteria for ARVC (Table 2), and histological evidence of ARVC was present at autopsy (Figure 3C).

**Phenotype Analysis**

Among the 7 families with TTN variants, a total of 14 individuals (9 male and 5 female individuals) were found to be carriers, with 12 meeting 2010 criteria for ARVC.23 Details of the phenotype features are reported in Table 2, and the pedigrees of the 7 kindreds are shown in Figure 1. Two of the 14 TTN variant carriers did not meet ARVC criteria: DNRVC008-III:4 and TSRVD001-IV:3, who had atrial fibrillation and conduction disease necessitating a pacemaker, indicating incomplete penetrance.

Mean age of onset of the TTN carrier cohort was 38 years, ranging from 19 to 59 years, thus indicating variability of expression. Most patients presented with symptoms related to ventricular arrhythmias (palpitations, n = 9; syncope, n = 7) and heart failure (dyspnea on exertion, n = 8). Five patients (36% of carriers) had evidence of left ventricular involvement with decreased left ventricular ejection fraction or enlargement on echocardiogram, and 9 (64%) showed progressive myocardial dysfunction (Table 2), leading to heart transplantation in 6 (43%) or causing death for progressive intractable heart failure at a mean of 55 years of age (range, 50 to 66 years; Table 2). Indications for heart transplant were intracardiac predominantly right heart failure (5 cases) and intracardiac ventricular arrhythmias (1 case). Two patients were resuscitated from cardiac arrest (DNRVD002-IV:3, and DN006-II:2). There was a history of sudden cardiac death in 5 of 7 families. The ECG showed epsilon waves in 3 cases, all belonging to the large kindred TSRVD001 (III-3, IV:4, and IV:7; Figure 4). Remarkably, TTN variant carriers had frequent conduction disease (11 of 14 patients in 6 families) in the course of their follow-up, leading to a permanent pacemaker in 8 of 14 patients (3 of 7 probands; Table 2). On the contrary, in the group of ARVC patients noncarriers of TTN variants, only 2 developed bradyarrhythmias requiring a pacemaker (2 of 31 probands, 2 of 50 affected family members; P < 0.001, Fisher exact test). Atrial arrhythmias (atrial fibrillation and atrial flutter) were also common (8 of 14 in 6 families) and were associated with right atrial dilation in 3 cases in whom measurement data were available.
Five patients received implanted defibrillators because of a history of ventricular arrhythmias. A detailed description of the phenotype of family TSRVD001 is reported in the online-only Data Supplement.

Table 2. Continued

<table>
<thead>
<tr>
<th>Protein Analysis</th>
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| The Thr2896Ile residue exchange substitutes a hydrophobic isoleucine for the hydrophilic threonine in the Ig10 immunoglobulin domain; sequence alignment with known Ig domain structures shows that the mutated residue is located near the A'β-strand of Ig10 (Figure 3A and 3B). Ig10 is found in the proximal tandem Ig repeat region near the Z disk, critical for the generation of passive cardiomyocyte tension. Steered molecular dynamics simulations have shown that the hydrogen bond network between parallel A'β and Gβ-strands is crucial for determining the mechanical stability of Ig domains, and atomic force microscopy experiments have shown that point mutations in A' strand residues can reduce the force needed to unfold Ig domains. Therefore, we hypothesized that the mutation near the A'β-strand in Ig10 reduces the mechanical stability of Ig10 and increases the probability of domain unfolding. To examine this experimentally, we produced the WT and the Thr2896Ile mutant version of Ig10 in E. coli. Expressed proteins were separated on 7% native tris-glycine polyacrylamide gels. The reduced electrophoretic mobility of mutant Ig10 that was found indicates a larger hydrodynamic radius in the mutant, suggestive of a less compact folded state (Figure 6A). To study this further, we determined next the intrinsic

Figure 4. An ECG from individual IV-7 from family TSRVD001 (Thr2896Ile). Arrows indicate epsilon waves.
fluorescence of the single tryptophan residue (W) of the domain, located in the hydrophilic core fold of Ig10, as a function of the concentration of the chemical denaturant guanidine chloride (GuCl). In the presence of 3 mol/L GuCl, the fluorescence levels of the 2 protein types were both low, indicating that W was exposed to a hydrophilic surrounding in a nonfolded state (Figure 6B). In the absence of denaturant, the fluorescence of the mutant Ig10 was lower than for WT Ig10, suggesting that the central W was less buried inside the Ig10 hydrophobic core (Figure 6B). We also applied heteronuclear single quantum correlation nuclear magnetic resonance to determine whether mutant Ig10 indeed has a less well-folded state. The $^{13}$C$^{15}$N-labeled mutant Ig10 exhibited 2 sets of nuclear magnetic resonance signals: 1 set with chemical shifts similar to those present in the WT domain and 1 set with less chemical shift dispersion (Figure 6C; spectra of WT [red] and mutant [blue] Ig10). Therefore, the mutant Ig10 domain coexists as a mixture of well-structured and less structured sequences in solution. Taken together, these data demonstrate that the mutant domain is less stable than the WT domain and is more likely to adopt an unfolded state. Finally, we also compared the susceptibility for proteolytic degradation of the WT and mutant titin peptides. Incubation of Ig7 to Ig13 spanning fragments with a low level of the protease trypsin was found to cause more prominent proteolysis in the sample containing the centrally located mutant Ig10 (Figure 6D and 6E).

Discussion

Titin is the largest protein known, and its 363 exons, located at chromosome position 2q31, span $\sim$0.3 Mb. Given the huge size of the titin gene, a large number of disease-causing mutations is expected, but compared with other much smaller sarcomeric proteins (eg, >100 mutations in the 15-times-smaller myosin heavy chain), relatively few titin mutations have been reported until now. Gerull et al described TTN mutations segregating with DCM, a 2-base insertion and a 1-bp deletion both causing a truncated A-band titin, and a Trp930Arg mutation in the Z disk. Itoh-Satoh et al and Matsumoto et al reported DCM TTN mutations with Arg743Val and Val54Met Z-disk region mutations at the site where titin interacts with several Z-disk proteins, Glu4053ter and Ser4465Asn mutations, which were hypothesized to alter the N2B cardiac-specific spring element, and an Arg25618Gly mutation mapping to the inextensible A band. The investigators suggested a decreased binding (loss of function) of mutant titin to titin-binding proteins. On the other hand, hypertrophic cardiomyopathy–causing mutations Arg25618Gly and Ser3799Tyr appear to increase binding to $\alpha$-actinin and FHL2, respectively (gain of function). Several mutations in and near the terminal exon of TTN cause titin myopathies that does not involve any apparent cardiac signs or symptoms. The mechanisms by which constitutively expressed titin mutations induce muscle type-specific myopathies are currently unknown and warrant future study.

In the present work, we sequenced titin in a cohort of ARVC families and discovered novel TTN variants in 18% of the families. The most prominent variant was Thr2896Ile, which showed strong segregation evidence for being pathogenic because it was present in 9 confirmed/obligate ARVC subjects, including 2 fifth-degree relatives, absent in 300 cardiomyopathy and 400 control chromosomes, and scored intolerant by SIFT and PolyPhen predictive algorithms. The phenotype of ARVC TTN variant carriers was characterized by progressive right ventricular dilatation and dysfunction, leading to death or cardiac transplantation. Right ventricular pathology, when available, showed fibrous or fibrofatty replacement. Common among TTN-ARVC families were reports of syncope and sudden unexplained death, which were often the presenting symptom in affected individuals. One
subject (TSRVD001-IV:3) who had normal biventricular function and did not meet full ARVC diagnostic criteria developed atrial fibrillation and early conduction system disease, findings more typical of advanced ARVC. This could suggest that conduction system disease and atrial arrhythmia could be early features of the TTN phenotype. Atrial fibrillation and conduction disease, which have been described previously in ARVC, were more common in our population among TTN variant carriers than noncarriers. We speculate that these phenotypic findings in ARVC might therefore implicate underlying TTN defects. The unusually high proportion of these findings in our study could suggest that there may be phenotypic overlap between ARVC and other conduction system disorders in carriers of TTN mutations. The most remarkable finding was the high prevalence of conduction disease in these patients, ranging from sinus node dysfunction to atrioventricular and intraventricular blocks, requiring a permanent pacemaker in >50% of affected carriers. These findings, along with the presence of atrial arrhythmia, suggest pathological changes beyond the right ventricular wall, possibly extending to the atrial myocardium. No evidence of involvement of other organs or systems was found in our cohort, as reported for nondesmosomal genes. Segregation data on the remaining 7 TTN variants were extremely limited in other families in our study, and although it is likely that some of these variants may represent pathogenic changes, to establish this conclusively requires future studies.

The Thr2896Ile variant appears to have a functional consequence, and on the basis of our in vitro data, we predict that at low sarcomere stretch when passive force is low, the mutant Ig10 is partially folded, and as sarcomere stretch increases and force rises, the likelihood of unfolding will exceed that of the WT domain. The effect of unfolding of a single domain on passive force will be small, considering that a large number of Ig domains (38 in N2B titin and 50 in N2BA titin) make up the extensible spring region, in addition to the N2B and PEVK spring elements. Using a serially linked wormlike chain model for simulating force-extension curves of titin, we calculated that, at a sarcomere length of 2.2 μm, the reduction is <3% of the total passive force (for details, see the study by Watanabe et al). This is a relatively small reduction, and it seems unlikely that force reduction caused by unfolding of a single domain initiates the disease pathology. Instead, the trypsin degradation experiments (Fig-
ure 6D and 6E) suggest that the mutant domain is more vulnerable to proteolysis and degradation and this increased vulnerability might be an essential step in the disease pathology. Unfolding of mutant Ig10 and the ensuing titin proteolysis will be most prominent in those regions of the heart where the spring elements of titin are most extended. The thin-walled stress-susceptible right ventricle might be such a region, especially at the transitional junction of the intercalated disk, where the strain of titin might far exceed that encountered elsewhere in the heart. Thus, we propose that titin mutations found in ARVC lower Ig domain stability and that this leads to titin degradation, which initiates the pathological process that eventually leads to ARVC. Future work is needed to test this hypothesized disease mechanism.

Conclusions

We discovered novel variants in the giant sarcomeric protein titin that are associated with ARVC. The phenotype of TTN variant carriers includes severe biventricular dysfuction, conduction disease, and sudden death. The Thr2896Ile ARVC TTN mutation has increased susceptibility to proteolysis, and we propose that this is a primary step in the disease pathology. Considering the critical role of titin in multiple processes, including increased calcium sensitivity with stretch, cell signaling, and protein turnover, there are multiple pathways by which titin proteolysis might trigger the ARVC disease pathology, warranting future follow-up research. The involvement of titin and sarcomeric dysfunction in ARVC further expands the causes of ARVC beyond desmosomal proteins. Interestingly, pathological changes with fibrofatty replacement of the myocardium were previously reported in non-ARVC patients with desmin, myosin heavy chain, and PRKAG2 mutations. Extra- desmosomal mutations can induce fibrofatty replacement suggesting a more expanded view of the triggers of this myocardial reparative process. Importantly, the discovery of titin mutations in ARVC might allow the design of novel therapeutic strategies such as the design of drugs that reduce proteolytic degradation or enhance Ig domain stability.

Sources of Funding

This work was supported by National Institutes of Health grants N01-HV-48194, RO1 HL69071, RO1 HL062881, MO1 #RR00051-1575, and 1K23HL67915-01A1; American Heart Association grants 0150453N and 0250271N; Muscular Dystrophy Association grant PN0007-056; and the Wilhelm-Müller Foundation.

Disclosures

None.

References


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*Circulation.* 2011;124:876-885; originally published online August 1, 2011;
doi: 10.1161/CIRCULATIONAHA.110.005405

*Circulation* is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
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Print ISSN: 0009-7322. Online ISSN: 1524-4539

The online version of this article, along with updated information and services, is located on the
World Wide Web at:
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Phenotype characteristics of family TSRVD001 carrying the Thr2896Ile *TTN* mutation

The proband (III-1) of family #1 (Thr2896Ile) presented at the age of 49 years with a history of syncope and biventricular heart failure. He had atrial fibrillation, ventricular tachycardia and required a pacemaker for bradyarrhythmias. An endomyocardial biopsy showed fibrous-fatty infiltration of right ventricular wall. He progressed to pump failure and died at the age of 66 years. His sister (III-3) presented with similar symptoms and severe right heart failure in addition to atrial fibrillation, ventricular tachycardia and third degree atrio-ventricular block requiring a pacemaker. Her ECG showed an epsilon wave. She died of progressive heart failure at the age of 58 years and had transmural fibro-fatty infiltration of the right ventricular myocardium at autopsy (Fig. 3C). A first cousin once-removed (IV-6) also presented with syncope and symptoms of right and left heart failure. The ECG showed atrial fibrillation, and atrio-ventricular block requiring a pacemaker. She was found to have ventricular tachycardia with left bundle branch morphology and on echocardiography diffuse hypokinesis dilatation and budging of the right ventricle and left ventricular dysfunction. An endomyocardial biopsy was significant for fatty-substitution of the right ventricular myocardium. She was transplanted for progressive heart failure at the age of 57 years. Following her diagnosis, other relatives (IV-4, IV-7, V-3) developed syncope and heart failure and fulfilled the criteria of ARVC. Relative IV-5 died suddenly at the age of 35. Relative IV-3, who carried of the Thr2896Ile mutation,
had atrial fibrillation and a history of coronary artery disease. He did not fulfill the criteria for ARVC and was considered as “unknown status”. Among the family ancestors there were three other unexplained sudden deaths (II-5, III-5, III-6) at ages 40, 63 and 26 years, and two other relatives had sudden deaths at unknown ages (II-1 and II-3).