Arrhythmogenic Right Ventricular Dysplasia/Cardiomyopathy

5

From Desmosome to Disease

Moniek G.P.J. Cox and Richard N.W. Hauer

5.1 Introduction

Arrhythmogenic Right Ventricular Dysplasia/Cardiomyopathy (ARVD/C) is a disease characterized by progressive fibrofatty replacement of primarily the *right ventricle* (RV).[1–3]

Affected individuals typically present between the second and fourth decade of life with *arrhythmias* originating from the RV. However, ARVD/C can also be the cause of *sudden death* already in adolescence, mainly in athletes.[4] From autopsy studies, it is known that already in young teenagers massive amounts of fibrofatty tissue can replace major parts of normal myocardium (Fig. 5.1).

The first series of ARVD/C patients was published in 1982, when it was called a disease in which "the right ventricular musculature is partially or totally absent and is replaced by fatty and fibrous tissue."[1] This disease was initially thought to be a defect in RV development, which is why it was first called "dysplasia." In the past 25 years, increased insight in the development of the disease as well as the discovery of pathogenic mutations involved, led to the current idea that ARVD/C is a genetically determined "cardiomyopathy."[3,5] The molecular genetic era has provided new insight in the understanding that ARVD/C is a desmosomal disease resulting from defective cell adhesion proteins. The first disease-causing gene, encoding the desmosomal protein *Plakoglobin* (*JUP*), was identified in patients with Naxos disease, an autosomal recessive variant of ARVD/C.[6] Its discovery pointed research in the direction of other desmosomal genes. Until 2004, evidence for genes underlying the autosomal dominantly inherited ARVD/C had been very limited, with three genes and six loci being identified.[7–15] The *Desmoplakin* gene (*DSP*) was the first desmosomal protein gene to be associated with the autosomal dominant form of ARVD/C.[15] It was followed by discovery of mutations in *Plakophilin-2* (*PKP2*), *Desmoglein-2* (*DSG2*), and *Desmocollin-2* (*DSC2*), also components of the cardiac desmosome.[16–18] Impaired desmosomal function may

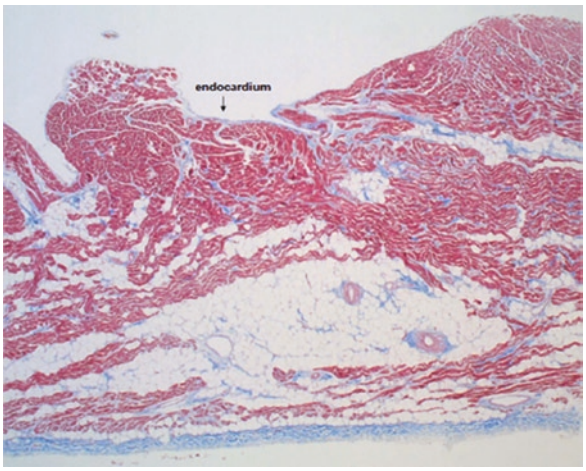

Fig. 5.1 Histology of right ventricle of a 13-year-old girl who died suddenly during exercise. AZAN staining (400×) with cardiac myocytes (red), collagen (blue), and adipocytes (*white*). Shown is the typical pattern of arrhythmogenic right ventricular dysplasia/cardiomyopathy (*ARVD/C*) with strands of fibrosis reaching all the way to the endocardium. Bundles of cardiac myocytes are embedded in between the fibrotic strands, particularly in the subendocardial layers. These interconnecting bundles of myocytes give rise to activation delay and re-entrant circuits, the typical electrophysiologic substrate for ventricular arrhythmias in ARVD/C. The large homogeneous subepicardial area of adiposites is not arrhythmogenic, is not typical for ARVD/C, and is also observed in the cor adiposum

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result in disruption of the myocardial architecture, leading to activation delay and thereby life-threatening arrhythmias. In a few rare cases, autosomal dominant ARVD/C has been linked to other genes unrelated to the cell adhesion complex, i.e., the genes encoding the cardiac ryanodine receptor (RyR2), the transforming growth factor-β3 gene (TGFβ3), and transmembrane protein 43 (*TMEM43*).[13, 14, 19]

With mutations found in about half of the patients, mainly in desmosomal genes and *PKP2* in particular, classical ARVD/C is currently considered a genetically determined desmosomal disease.

This chapter will give an overview of ARVD/C, starting from the genetic defects and via the pathophysiologic mechanism to clinical diagnosis, treatment, and prognosis.

5.2 Molecular and Genetic Background

5.2.1 Desmosome Function

The functional and structural integrity of cardiac myocytes is enabled by cell adhesion junctions in the *intercalated disk*. Intercalated disks are located between cardiomyocytes at their longitudinal ends and contain three different kinds of intercellular connections: desmosomes, adherens junctions, and gap junctions.

Desmosomes are important for cell–cell adhesion and are predominantly found in tissues that experience mechanical stress: the heart and epidermis. They couple cytoskeletal elements to the plasma membrane at cell–cell adhesions. Desmosomes also protect the other components of the intercalated disk from mechanical stress and are involved in structural organization of the intercalated disk. Desmosomes consist of multiple proteins, which belong to three different families:

1. Transmembranous cadherins: desmogleins and desmocollins
2. Linker armadillo repeat proteins: plakoglobin and plakophilin
3. Plakins: desmoplakin and plectin

Figure 5.2 schematically represents the organization of the various proteins in the cardiac desmosome.

Within desmosomes, cadherins are connected to armadillo proteins, which for their parts interact with plakins. The plakins anchor the desmosomes to intermediate filaments, mainly desmin. Thereby, they form a three-dimensional scaffold providing mechanical support.

Adherens junctions act as bridges that link the actin filaments within sarcomeres of neighboring cells. These junctions are involved in force transmission and together with desmosomes, these mechanical junctions act as "spot welds" to create membrane domains that are protected from shear stress caused by contraction of the neighboring cells. Furthermore, they facilitate

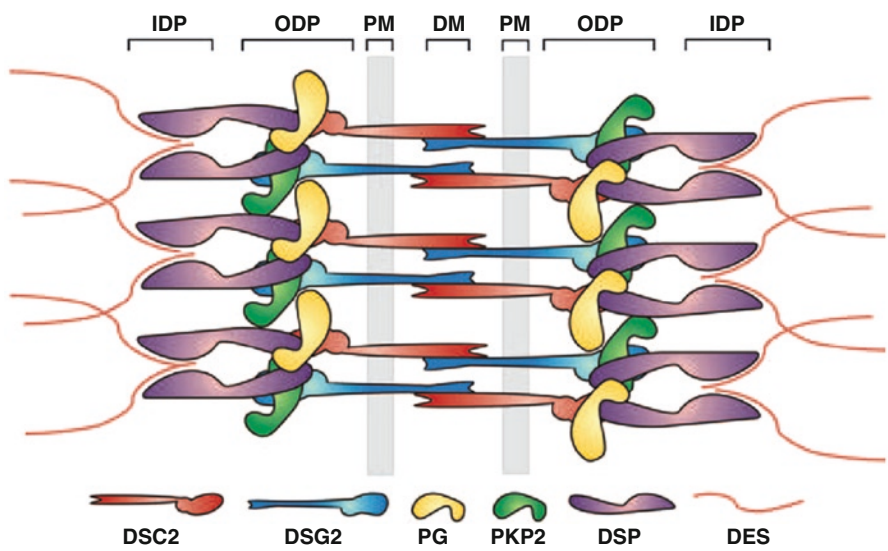

Fig. 5.2 Schematic representation of the molecular organization of cardiac desmosomes. The plasma membrane (*PM*) spanning proteins Desmocollin-2 (*DSC2*) and Desmoglein-2 (*DSG2*) interact in the extracellular space at the dense midline (*DM*). At the cytoplasmic side, they interact with plakoglobin (*PG*) and PKP2 at the outer dense plaque (*ODP*). PKP2 and PG interact also with Desmoplakin gene (*DSP*). At the inner dense plaque (*IDP*), the C-terminus of DSP anchors the intermediate filament desmin. (Reprint with permission from Van Tintelen et al. Curr Opin Cardiol 2007[27])

assembly and maintenance of gap junctions, securing intercellular electrical coupling.

Cardiomyocytes are individually bordered by a lipid bilayer, which gives a high degree of electrical insulation. The electrical current that forms the impulse for mechanic contraction can travel from one cell to the other via gap junctions. Gap junctions provide electrical coupling by enabling ion transfer between cells. The number, size, and distribution of gap junctions all influence impulse propagation in cardiac muscle. Consequently, alterations in function or structure of gap junctions can lead to intercellular propagation disturbances and contribute to arrhythmogenesis.[20]

Thus, the intercalated disk is an intercellular structure, where desmosomes and adherens junctions not only provide mechanical strength, but also protect the interspersed gap junctions, enabling electrical coupling between cells.

5.2.2 Desmosomal Dysfunction and ARVD/C Pathophysiology

Although the functions of different parts of the intercalated disk seem clear, the exact mechanism through which the mutations of desmosomal protein genes exactly cause disease remains to be elucidated. Various hypotheses, all based on the different functions of desmosomes, have been proposed.

First of all, genetic defects in a desmosomal protein are thought to lead to impairment in mechanical function provoking detachment of myocytes at the intercalated disks, particularly under condition of mechanical stress (like that occurring during competitive sports activity). Such defective mechanical connection followed by mechanical and electrical uncoupling of cardiomyocytes leads to cell death with fibrofatty replacement. Interconnecting bundles of surviving myocardium embedded in the fibrofatty tissue lead to lengthening of conduction pathways (Figure 5.1), and load mismatch. This results in marked *activation delay*, which is the pivotal mechanism for re-entry and thereby ventricular tachycardia (VT). Previous invasive electrophysiologic studies have, by various mapping techniques, confirmed that VT in patients with ARVD/C is due to re-entry circuits in areas of abnormal myocardium.[21] In this structural model, environmental factors such as exercise or inflammation from viral infection could aggravate impaired adhesion and accelerate disease progression. The right ventricle might be more vulnerable to disease than the left because of its thinner walls and its normal dilatory response to exercise.

Second, recent studies have shown that impairment of cell–cell adhesion due to changes in desmosomal components may affect amount and distribution of other intercalated disk proteins, including connexin43, the major protein forming gap junctions in the ventricular myocardium.[22–24] This was shown for DSP and JUP by Western blotting and confocal immunofluorescence techniques, but alterations in other desmosomal components such as PKP2, DSG2, and DSC2 are thought to have similar effects. Changes in number and function of gap junctions will diminish intercellular electrical coupling. This may contribute to intraventricular activation delay, and the substrate for re-entry.

The third hypothesis involves the canonical Wnt/β-catenin signaling pathway. Plakoglobin can localize both to the plasma membrane and the nucleus. It was demonstrated that disruption of desmoplakin frees plakoglobin from the plasma membrane allowing it to translocate to the nucleus and suppress canonical Wnt/β-catenin signaling. Wnt signaling can inhibit adipogenesis by preventing mesodermal precursors from differentiating into adipocytes.[25] Suppression of Wnt signaling by plakoglobin nuclear localization could, therefore, promote the differentiation to adipose tissue in the cardiac myocardium in patients with ARVD/C.[26]

Finally, since ion channels, like the Na^+ channel, are also located in the intercalated disk, they might be disrupted and contribute to arrhythmogeneity as well, although at this point this is hypothetical.

The pathophysiological mechanisms proposed above are not mutually exclusive and could occur at the same time.

5.2.3 Desmosomal Disease

Two patterns of inheritance have been described in ARVD/C. The most common or classical form of ARVD/C is inherited as an autosomal dominant trait. The rare *Naxos disease* and *Carvajal syndrome* are inherited autosomal recessively. Table 5.1 summarizes the different genes involved in ARVD/C with the corresponding phenotypes.

Table 5.1 Mutated genes with concurrent type of arrhythmogenic right ventricular dysplasia/cardiomyopathy (*ARVD/C*) (Modified from Van Tintelen et al. Curr Opin Cardiol 2007[27])

	Gene	Type of disease	Inheritance trait
Desmosomal	PKP2	Typical ARVD/C	Autosomal dominant
	DSG2	Typical ARVD/C	Autosomal dominant
	DSC2	ARVD/C	Autosomal dominant
	JUP	Naxos disease	Autosomal recessive
	DSP	Carvajal syndrome	Autosomal recessive
		ARVD/C	Autosomal dominant
		LDAC	Autosomal dominant
Nondesmosomal	RyR2	CPVT	Autosomal dominant
		ARVD/C	Autosomal dominant
	TGF-β	Typical ARVD/C	Autosomal dominant
	TMEM43	ARVD/C	Autosomal dominant

CPVT catecholaminergic polymorphic VT, *LDAC* left dominant arrhythmogenic cardiomyopathy. See text for other abbreviations

5.2.4 Autosomal Recessive Disease

In Naxos disease, the affected individuals were found to be homozygous for a 2 base pair deletion in the JUP gene.[6] All patients who are homozygous for this mutation have diffuse palmoplantar keratosis and woolly hair in infancy; children usually have no cardiac symptoms, but may have electrocardiographic abnormalities and nonsustained ventricular arrhythmias.[28] In an Arab family, an autosomal recessive mutation in the desmoplakin gene caused ARVD/C, also combined with woolly hair, and a pemphigus-like skin disorder.[29] A different autosomal recessive disease, Carvajal syndrome, is also associated with a desmoplakin gene mutation, and is manifested by woolly hair, epidermolytic palmoplantar keratoderma, and cardiomyopathy.[30] The cardiomyopathy of Carvajal syndrome was thought to have a predilection for the left ventricle, but subsequent evaluation of a deceased child revealed typical ARVD/C changes in both ventricles.[24] The cardiac phenotype in the Arab family appeared to be classic ARVD/C.

5.2.5 Autosomal Dominant Disease

Mutations in the gene encoding the intracellular desmosomal component desmoplakin lead to "classic ARVD/C" with a clinical presentation of VT, sudden death, and LV involvement as the disease progresses.[15,31,32] Desmoplakin gene mutations have also been associated with predominantly left-sided ARVD/C and, as noted above, with autosomal recessive disease.

Overall, mutations in the *PKP2* gene are the most frequently observed in ARVD/C. Figure 5.3 shows the pedigree of a family with a *PKP2* mutation. Incomplete penetrance and clinical variability is well documented.

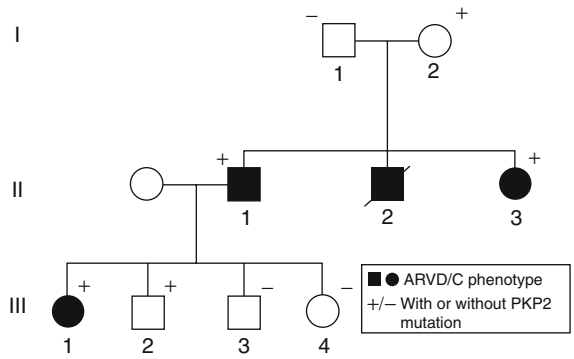

Fig. 5.3 Pedigree of family with arrhythmogenic right ventricular dysplasia/cardiomyopathy (*ARVD/C*) and plakophilin-2 (*PKP2*) mutation. This figure shows the variability in penetrance and clinical expression. Both the 72-year-old grandmother (I:2) and 20-year-old grandson (III:2) are free of any signs of disease, despite carrying the mutation. The proband (II:1) was resuscitated at age 35, his brother (II:2) died suddenly at age 18. Both the proband's sister (II:3) and daughter (III:1) were diagnosed with the disease due to a positive family history, and RV structural abnormalities. The sister (II:3) of the proband has structural and ECG abnormalities, but no arrhythmias

In four studies from different countries, analyzing 56 to 100 ARVD/C patients each, the following observations were made.[16, 33–35] *PKP2* mutations were found in 11–43% of unrelated index patients who fulfilled diagnostic Task Force criteria (TFC) for ARVD/C. In the current Dutch ARVD/C cohort, 67 of 125 (54%) probands carry a pathogenic *PKP2* mutation. In total, 21 different mutations have been observed: 10 nonsense, 4 missense, 3 frameshifts, 2 at splice sites, and 2 deletions. Haplotype analysis previously performed suggested that founder mutations were responsible for 4 of the 14 different mutations identified.[35] Among index patients with a positive family history of ARVD/C, 70% had a *PKP2* mutation.[35] No specific genotype–phenotype correlations were established, except that patients with *PKP2* mutation presented at a younger age (28 vs. 36 years), and negative T waves in V1-3 occurred more often in *PKP2* mutation carriers. Thus, *PKP2* appears to be a relatively commonly mutated gene in ARVD/C patients, particularly in cases with a documented family history. However, there does not appear to be a substantial phenotypic distinction from other mutations, and due to relatively small sample sizes and the potential for referral bias, the incidence of *PKP2* mutations in a broader population may be lower.

Because of the known association of ARVD/C with defects in other desmosomal proteins, a series of patients with ARVD/C were screened for mutations in the gene encoding the transmembranous desmosomal component DSG2 as well.[17] Among 80 unrelated probands, 26 were known to have *DSP* or *PKP2* mutations. Direct sequencing of *DSG2* in the other 54 patients revealed nine distinct mutations in eight individuals. These individuals demonstrated typical clinical characteristics of ARVD/C. An analogous study of 86 ARVD/C probands identified eight novel *DSG2* mutations in nine probands. Clinical evaluation of family members with *DSG2* mutations revealed penetrance of 58% using *Task Force criteria* and 75% using proposed modified criteria.[36] Morphological abnormalities of the right ventricle were present in 66% of gene carriers, LV involvement in 25% and classical right precordial T-wave inversion in only 26%. The authors noted that disease expression of DSG2 mutations was of variable severity, but that overall penetrance was high and LV involvement prominent.[37]

In the gene encoding DSC2, another important transmembranous desmosomal cadherin, two heterozygous mutations (a deletion and an insertion) were identified in 4 of 77 probands with ARVD/C.[18] The identification of the fifth desmosomal cell adhesion gene abnormality further supports the hypothesis that ARVC is usually a disease of cell adhesion.

5.2.6 Other, Nondesmosomal, Genes

Mutations in the gene encoding the cardiac ryanodine receptor RyR2, which is responsible for calcium release from the sarcoplasmic reticulum, have been described in only one Italian ARVD/C family.[13] Affected patients have exercise-induced polymorphic VT.[38] Mutations in *RyR2* have primarily been associated with familial catecholaminergic polymorphic VT without ARVD/C.[19, 39] RyR2 mediates the release of calcium from the sarcoplasmic reticulum that is required for myocardial contraction. The FK506 binding protein (FKBP12.6) stabilizes RyR2, preventing aberrant activation. The mutations in *RyR2* interfere with the interaction with FKBP12.6, increasing channel activity under conditions that simulate exercise.[39] Although the general opinion is that *RyR2* mutations lead to catecholaminergic polymorphic VT, without structural abnormalities, the mutations in ARVD/C have been advocated to act differently from those in familial polymorphic VT without ARVD/C.[40–42]

Transforming growth factor-β-3 (TGFβ3) regulates the production of extracellular matrix components and modulates expression of genes encoding desmosomal proteins. Its gene has been mapped to chromosome 14. Sequencing studies failed to identify any disease-causing mutations in the exonic regions of *TGFβ3*. This led to screening of the promoter and untranslated regions, where a mutation of the *TGFβ3* gene was found in all clinically affected members of a large family with ARVD/C.[14] The mutation is predicted to produce an amino acid substitution in a short peptide with an inhibitory role in TGFβ3 regulation. The implication of these observations is that regulatory mutations resulting in overexpression of TGFβ3 may contribute to the development of ARVD/C in these families. The TGFβ family of cytokines stimulates production of components of the extracellular matrix. It is therefore possible that enhanced TGFβ activity can lead to myocardial fibrosis. However, genetic analysis of two other families with ARVD/C failed to identify mutations in any of the regions of the TGFβ3 gene.

A missense mutation in the *TMEM43* gene was found in 15 unrelated ARVD/C families from a genetically isolated population in New Foundland and caused a fully penetrant, sex-influenced, high-risk form of ARVD/C.[19] The *TMEM43* gene contains the response element for PPAR gamma, an adipogenic transcription factor. The *TMEM43* gene mutation is thought to cause dysregulation of an adipogenic pathway regulated by PPAR gamma, which may explain the fibrofatty replacement of myocardium in ARVD/C patients.

5.3 Epidemiology

Estimations of the prevalence of ARVD/C in the general populations vary from 1:2,000 to 1:5,000.[43] The exact prevalence of ARVD/C, however, is unknown and is possibly higher because of the existence of many nondiagnosed or misdiagnosed cases.

The disease appears to be especially common in adolescents and young adults in northern Italy, accounting for approximately 11% of cases of sudden cardiac death overall and even 22% in athletes.[44, 45] In as many as 20% of sudden deaths occurring in people under 35 years of age, features of ARVD/C were detected at postmortem evaluation.[45] In nearly half of them, no prior symptoms had been reported. In contrast, ARVD/C has rarely been diagnosed in the United States. Founder mutations have been identified, e.g. in the Netherlands, and these could in part explain the difference in prevalence in different geographical areas.

ARVD/C has a reduced penetrance and extremely variable clinical expression. For instance, family screening has identified pathogenic mutation carriers, who had stayed free of any sign of disease up to over 70 years of age (Fig. 5.3).

Although from a genetic point of view, both men and women have to be equally affected, men are more frequently diagnosed with ARVD/C than women, with an approximate ratio of 3:1. However, as many women as men do show at least some signs of disease, but women more often do not fulfill enough criteria to meet the diagnosis. Factors explaining this difference in severity of disease expression have not yet been elucidated. It is speculated that (sports) activity or hormonal factors may play a role. A familial background has been demonstrated in >50% of ARVD/C cases.

5.4 Clinical Presentation

ARVD/C patients typically present between the second and fourth decade of life with VT originating from the right ventricle. However, in a minority of cases sudden death, possibly at a young age, or RV failure are the first signs. Based on clinicopathologic and patient follow-up studies, four different disease phases have been described for the classical form of ARVD/C, i.e., primarily affecting the RV (Table 5.2).

1. Early ARVD/C is often described as "concealed" owing to the frequent absence of clinical findings, although minor ventricular arrhythmias and subtle structural changes may be found. Although patients tend to be asymptomatic, they may nonetheless be at risk of sudden death, mainly during intense exercise.
2. The overt phase follows, in which patients suffer from palpitations, syncope, and ventricular arrhythmias of left bundle branch block morphology, ranging from isolated ventricular premature complexes to sustained VT and ventricular fibrillation (VF).
3. The third phase is characterized by RV failure due to progressive loss of myocardium with severe dilatation and systolic dysfunction, in the presence of preserved LV function.
4. Biventricular failure occurs, due to LV involvement. This phase may mimic dilated cardiomyopathy (DCM) and may require cardiac transplantation.

In the initially described classical form of ARVD/C, the RV is primarily affected with possibly (in a later stage) some LV involvement. Two additional distinct

Table 5.2 Different phases of disease severity

Phase	Characteristics
1. Concealed	Asymptomatic patients with possibly only minor ventricular arrhythmia and subtle structural changes However, risk of sudden death
2. Overt	Symptoms due to LBBB VT or multiple premature complexes, with more obvious structural RV abnormalities
3. RV failure	With relatively preserved LV function
4. Biventricular	Significant overt LV involvement

patterns of disease have been identified by clinicogenetic characterization of families. These are the left dominant phenotype, with early and predominant LV manifestations, and the biventricular phenotype with equal involvement of both ventricles. Recent immunohistochemical analysis of human myocardial samples demonstrated that on a desmosomal level, both ventricles are affected by the disease.[73] A marked reduction in immunoreactive signal levels for Plakoglobin was observed both in the right and left ventricle, independent of genotype. This strengthens the idea that in essence, ARVD/C is a biventricular disease. However, histologically and functionally overt manifestations of the disease usually start in the RV. The reason for this is still unclear. The most advocated idea is that the thin-walled RV is less able to withstand pressure (over)load when the mechanical junctions have an impaired function.

5.5 Clinical Diagnosis

Diagnosis of ARVD/C can be very challenging and can only be made when all other diseases causing VT episodes from the RV have been ruled out (see paragraph on differential diagnosis). Although VF and sudden death may be the first manifestations of ARVD/C, symptomatic patients typically present between age 20 and 40 years with sustained VT with left bundle branch block morphology, thus originating from the RV. The occurrence of VT episodes is usually driven by adrenergic stimulation and starts mainly during exercise, especially competitive sports. ARVD/C is a disease that shows progression over time, and may manifest differently according to the time of patient presentation.

The gold standard for ARVD/C diagnosis is the demonstration of transmural fibrofatty replacement primarily of right ventricular myocardium, determined at surgery or postmortem. Predilection sites for these structural abnormalities are the so-called triangle of dysplasia formed by the RV outflow tract (RVOT), the apex, and the subtricuspid region. In daily clinical practice, this definition of diagnosis is not usable. Even endomyocardial biopsies have major limitations. Tissue sampling from the affected often thin RV-free wall, directed by imaging techniques or voltage mapping, is rather straightforward, but is associated with a risk of perforation. Sampling from the interventricular septum is relatively safe. However, the septum is histopathologically rarely affected in ARVD/C. In addition, even in potentially affected areas, histology may be classified as normal because of the segmental nature of the lesions. Finally, since subendocardial layers are usually not affected in an early stage of the disease, histologic diagnosis may be hampered by the nontransmural nature of endomyocardial biopsies.[46, 47]

Clinical diagnosis has been facilitated by a set of clinically applicable criteria for ARVD/C diagnosis defined by a Task Force in 1994.[48] Based on the evidence available at that time, the Task Force included six different groups of clinical criteria. Within these groups, diagnostic criteria were assigned major or minor according to their specificity for the disease. Every major criterion is scored as two points and every minor as one point. In total, four points have to be scored in order to fulfill the ARVD/C diagnosis, i.e., two major, one major plus two minor, or four minor criteria. From each different group, only one criterion can be counted for diagnosis, even when multiple criteria in one group are being fulfilled. Recently, these 1994 Task Force criteria have been revised. See Table in Addendum.

Specific tests are recommended in all patients suspected of ARVD/C. A 12-lead ECG, signal averaged ECG (SAECG; when available), 24 h Holter monitoring, exercise testing, and 2D-echocardiography should be performed in all. When appropriate, more detailed analysis of the RV can be done by cardiac MRI or computed tomography. Eventually, invasive tests are also available for diagnostic purposes: endomyocardial biopsy, RV cine-angiography, and electrophysiologic testing.

Table 5.3 gives an overview of the Task Force Criteria (TFC).

5.6 ECG Criteria

Criteria on ECG changes have to be obtained in normal sinus rhythm and while off anti-arrhythmic drugs. Furthermore, a complete right bundle branch block has to be absent. ECG changes are detected in up to 90% of ARVD/C patients.

5.6.1 Depolarization Abnormalities

As explained above, RV activation delay is a hallmark of ARVD/C. This delay is conveyed by the criteria of

Table 5.3 Diagnostic Task Force Criteria[48]

Factor	Major criteria	Minor criteria
Family History	Familial disease confirmed at necropsy or surgery	Family history of premature sudden death (age <35 years) due to suspected ARVD/C Family history (clinical diagnosis based on present criteria)
ECG depolarization/conduction abnormalities	Epsilon waves or localized prolongation (>110 ms) of QRS complex in leads V_1–V_3	Late potentials on signal-averaged ECG
ECG repolarization abnormalities		Inverted T waves in right precordial leads (V_2 and V_3) in people aged >12 years and in absence of right bundle branch block
Arrhythmias		VT with LBBB morphology >1,000 premature ventricular complexes in 24 h on Holter monitoring
Global or regional dysfunction and structural alterations	RV akinetic or dyskinetic areas with diastolic bulging; Severe dilatation and reduction of RV ejection fraction with no or mild LV involvement (RV > LV)	Mild global RV dilatation or ejection fraction reduction with normal LV Mild segmental dilatation of RV
Tissue characteristics of walls	Fibrofatty replacement of myocardium on endomyocardial biopsy	

epsilon waves or localized prolongation of the QRS complex in V1-3 (>110 ms) and late potentials on SAECG.

Epsilon waves are defined as low amplitude potentials after and clearly separated from the QRS complex, in at least one of V1-3 (Fig. 5.4).[49] This highly specific major criterion is unfortunately observed in only a small minority of patients.[50, 51]

Localized prolongation of the QRS complex in V1-3 >110 ms is also a major diagnostic criterion. In one study, this was observed in as many as 70% of patients. However, most reports give lower percentages.[52]

The detection of *late potentials* on SAECG is the surface counterpart of delayed activation or late potentials detected during endocardial mapping in electrophysiologic studies. They are frequently found in patients with documented VT. However, these late potentials can also be observed after myocardial infarction and other structural heart diseases, and due to this lack of specificity were considered a minor criterion.

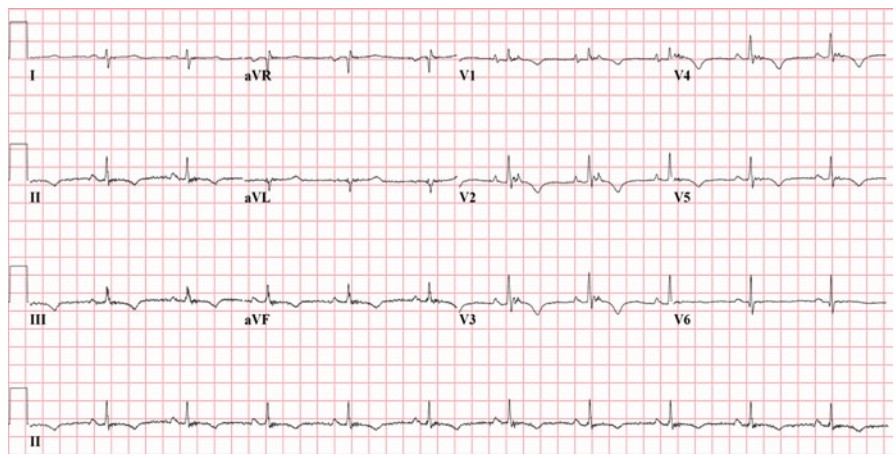

Fig. 5.4 Epsilon waves and negative T waves in V1-5

For all criteria on depolarization abnormalities, it is apparent that their finding will correlate with disease severity. For instance, a positive correlation has been found between late potentials and the extent of RV fibrosis, reduced RV systolic function, and significant morphological abnormalities on imaging.[53–55]

5.6.2 Repolarization Abnormalities

Negative T waves in leads V1 up to and including V3 form the minor ECG criterion on repolarization abnormalities (see Fig. 5.4). They are the most frequently observed criterion. In the initial series reported by Marcus et al., this was detected in over 85% of cases.[1] Subsequent studies have reported variable prevalences of right precordial T wave inversion, ranging from 19% to 94%.[48–52] The lower rates are often due to evaluation of family members, while higher rates are seen in series consisting of unrelated index patients. *T wave inversion* can be a normal feature of the ECG in children and early adolescence. Therefore, this finding is not considered pathogenic in persons aged 12 years and younger. Similarly, negative T waves are not to be judged in the presence of a right bundle branch block.

Although these negative T waves were observed consistently in a series of evaluated ARVD/C patients, T wave inversions in the right precordial leads can also be observed in 1–3% of the healthy populations aged 19–45 years, and in patients with right ventricular overload, such as major pulmonary embolism and intracardial left to right shunt, or may develop following intracranial hemorrhage as a sign of adrenergic response to the cerebral insult. Owing to this presumed lack of specificity, T wave inversions were included as a minor criterion.

5.6.3 Arrhythmias

Ventricular arrhythmias range from premature ventricular complexes to sustained VT and VF, leading to cardiac arrest.[56,57] Because of the origin in the RV, QRS complexes of ventricular arrhythmias show a left bundle branch block morphology. Moreover, the QRS axis gives an indication of the VT origin, i.e., superior axis from the RV inferior wall or apex and inferior axis from the RV outflow tract (see Fig. 5.5). Patients with extensively affected RV may show multiple VT morphologies.

VF is the mechanism of instantaneous sudden death especially occurring in young people and athletes with ARVD/C, who were often previously asymptomatic. In this subset of patients, VF may occur from deterioration of monomorphic VT, or in a phase of acute disease progression, due to acute myocyte death and reactive inflammation.[3]

5.6.4 Global and/or Regional Dysfunction and Structural Alterations

Evaluation of RV size and function can be done by various imaging modalities, including echocardiography, magnetic resonance imaging (MRI), computed

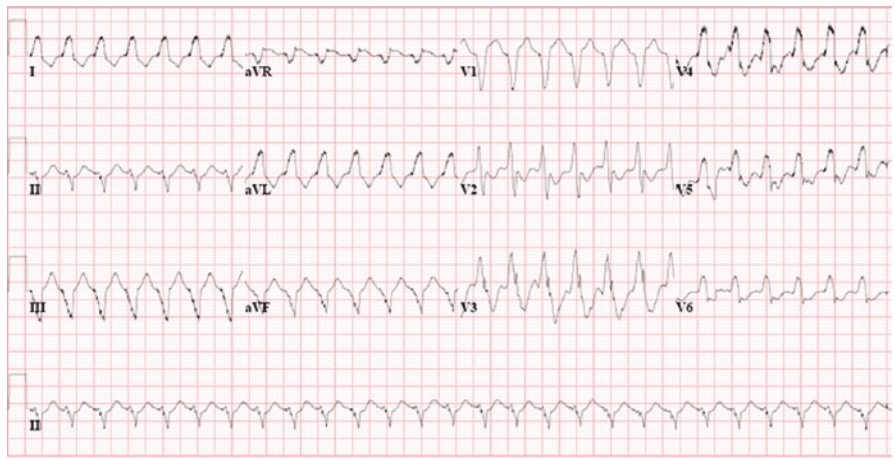

Fig. 5.5 ECG (25 mm/s) from arrhythmogenic right ventricular dysplasia/cardiomyopathy (*ARVD/C*) patient with plakophilin-2 (*PKP2*) mutation. This ventricular tachycardia (*VT*) has an LBBB morphology and superior axis, thus originates inferiorly from the right ventricle (*RV*)

tomography, and/or cine-angiography. According to the Task Force criteria, major criteria are defined as presence of large a- or dyskinetic areas in the RV or severe dilatation of the RV (RV larger than LV).[48] Less severe abnormalities are considered minor criteria. The Task Force criteria are unspecific, in the sense that no definitions of severe compared with less severe had been formulated. Although each of the different imaging modalities mentioned can detect severe structural abnormalities, the diagnostic value of each is less certain when evaluating mild cases of the disease. RV cine-angiography has historically been considered the gold standard to visualize RV structural abnormalities, with a high specificity of 90%.[58] Compared to cine-angiography, the noninvasive technique of echocardiography is widely used and serves as the first-line imaging technique in evaluating patients suspected of ARVD/C and in family screening. Especially with improvement of echocardiographic modalities, like three-dimensional echocardiography, strain and tissue Doppler, sensitivity and specificity of echocardiography have increased in the past years. Cardiac MRI is advantageous, since it has the unique possibility of visualizing the myocardium to characterize tissue composition, by differentiating fat from fibrous tissue by delayed enhancement. However, this technique is expensive and not widely available and requires great expertise to prevent mis- or overdiagnosis of ARVD/C.[59] Also, in ICD-carrying patients, this technique cannot be applied. Cardiac MRI has appeared to be the most common cause of overdiagnosis and physicians should therefore be very reluctant to diagnose ARVD/C when structural abnormalities are only present on MRI.[60,61] Furthermore, it is important to note that the presence of fat in the epi- and midmyocardial layers (without fibrosis) is known as cor adiposum and should not be considered diagnostic of ARVD/C.

5.6.5 Endomyocardial Biopsy

For reasons outlined earlier, undirected endomyocardial biopsies are rarely diagnostic. However, it had been included as a major criterion by the Task Force, since the finding of fibrofatty replacement was considered to strongly support any findings derived from other clinical investigations. The rather vague description of any "fibrofatty replacement of myocardium," had been further studied and specified by the pathology department of Padua, where histomorphometric parameters of myocytes, interstitium, fibrous tissue, and fatty tissue were evaluated. Diagnostic values indicating arrhythmogenic right ventricular cardiomyopathy were presence of <45% of cardiomyocytes, >40% fibrous tissue, and >3% of fatty tissue with 67% sensitivity and 91% specificity for at least one parameter.[62]

5.6.6 Family History

Already before the discovery of pathogenic mutations underlying the disease, it was recognized that ARVD/C often occurs in multiple members of the same family. Having a family member with proven ARVD/C was considered an increased risk for other family members to get the disease as well, and was therefore included as a minor diagnostic criterion. Also, sudden death of a family member under the age of 35 years, presumably but not proven to be due to ARVD/C-related arrhythmias, is a minor criterion. Pathologic confirmation of transmural fibrofatty replacement of the RV at autopsy or after surgical resection is the gold standard for ARVD/C diagnosis. Therefore, if this is diagnosed in a relative, it is considered a major criterion in the diagnosis of their relatives.

5.6.7 Discussion on Diagnostic Criteria

The current TFC are the essential standard for classification of individuals suspected of ARVD/C. In addition, its universal acceptance contributed importantly to unambiguous interpretation of clinical studies and facilitated comparison of their results. However, past years have shown that this set of criteria is very specific, but lacks sensitivity. From studies on larger numbers of patients and their family members, more insight has been gained into the disease. This leads to the possibility of various modifications and extensions by multiple groups.

A number of additional ECG markers have been reported, mainly reflecting right ventricular activation delay, including QRS and QT dispersion, parietal block (QRS duration in V1–V3 exceeds the QRS duration in V6 by >25 ms), localized right precordial QRS

prolongation (QRS duration in leads V1 + V2 + V3/V4 + V5 + V6 ≥ 1.2). Recently, Nasir et al. demonstrated that a prolonged (≥55 ms) S-wave upstroke in the right precordial leads correlates with disease severity and induction of VT during electrophysiological studies.[63] Our group improved this criterion and introduced prolonged *terminal activation duration* (*TAD*).[57] Prolonged TAD is defined as ≥ 55 ms, from the nadir of the S wave to the end of all depolarization deflections in V1-3, thereby covering all forms of RV activation delay, including epsilon waves (Fig. 5.6). This new criterion appeared to be more sensitive than all accepted and proposed diagnostic criteria on activation delay. Whereas other criteria were observed only in a minority, prolonged TAD was observed in as many as 71% of patients studied. Furthermore, we observed that VT with LBBB morphology and superior axis, and/or multiple different VT morphologies were recorded in 67% and 88% of ARVD/C patients, respectively. All three newly proposed additional criteria were highly specific for ARVD/C as well. However, since most markers have been deducted from ARVD/C-positive patients selected on the basis of the TFC, the additional diagnostic value still has to be evaluated in further prospective studies.

Multiple groups attempted to provide better quantification of normal and abnormal values of RV dimensions. Both for echocardiography and MRI, right ventricular dimensions of ARVD/C patients have been compared to controls.[64,65] Normal and cut-off values have been established based on standard deviations, as was done by the initial Task Force as well. The disadvantage of this method is that the value at two standard deviations from normal do not always represent the cutoff point optimal with respect to sensitivity and specificity.

Currently, a modification of the complete set of the 1994 TFC is being created. The new TFC will differ from the current ones, by including prolonged TAD and electrical axis of VT, using quantification of RV dimensions and have a role for results of genetic screening (see Table in Addendum).

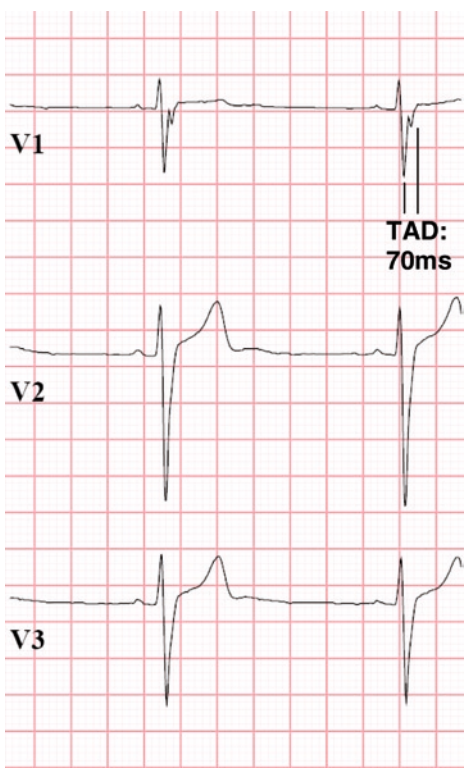

Fig. 5.6 Prolonged terminal activation duration (≥55 ms from nadir of S wave to end of depolarization)

5.6.8 Modifications for Family Members

The same set of diagnostic TFC is applied whether it concerns a proband or one of his family members. Based on the experience of similar criteria in diagnosing hypertrophic and dilated cardiomyopathies showing that many relatives have phenotypic abnormalities that, although "nondiagnostic," are nevertheless indicative of disease, a new set of TFC was proposed for family members of ARVD/C patients.[36] Hamid et al. reported that by using current TFC, familial disease was observed in 28% of index patients.[36] A further 11% of their relatives had minor cardiac abnormalities, which, in the context of an autosomal dominant disease, are likely to represent early or mild disease expression. They proposed that first-degree relatives of patients with proven ARVD/C with a positive SAECG, or a minor ECG, Holter, or echocardiographic abnormality from the present diagnostic criteria should get ARVD/C diagnosis as well. Furthermore, they advocate that the frequency of ectopic activity accepted as a marker of disease expression should be reduced from 1,000 to 200 ventricular premature complexes over a 24-h period. Hereby, they expect that disease classification of family members will be improved. However, because of the positive family history, family members will already fulfill one of TFC, either major or minor,

depending on the type of history. Therefore, they have to fulfill fewer of the other criteria to get the ARVD/C diagnosis. This can possibly lead to family members being stigmatized unnecessarily, since having a positive family history indicates a higher risk to develop ARVD/C, but does not change the degree to which they are actually affected by the disease.

5.6.9 Nonclassical ARVD/C Subtypes

5.6.9.1 Naxos Disease

All patients who homozygously carry the recessive JUP mutation for Naxos disease have diffuse palmoplantar keratosis and woolly hair in infancy; children usually have no cardiac symptoms, but may have ECG abnormalities and nonsustained ventricular arrhythmias.[23, 28] The cardiac disease is 100% penetrant by adolescence, being manifested by symptomatic arrhythmias, ECG abnormalities, right ventricular structural alterations, and LV involvement. In one series of 26 patients followed for 10 years, 62% had structural progression of right ventricular abnormalities, and 27% developed heart failure due to LV involvement.[28] Almost half of the patients developed symptomatic arrhythmias and the annual cardiac and SCD mortality were 3% and 2.3%, respectively, which are slightly higher than seen in autosomal dominant forms of ARVD/C. A minority of heterozygotes have minor ECG and structural changes, but clinically significant disease is not present.

5.6.9.2 Carvajal Syndrome

Carvajal syndrome is associated with a *DSP* gene mutation, and is also a recessive disease manifested by woolly hair, epidermolytic palmoplantar keratoderma, and cardiomyopathy.[30] All patients diagnosed so far came from Ecuador. The cardiomyopathy part of Carvajal syndrome was first thought to be mainly left ventricular, with dilated left ventricular cardiomyopathy. A number of the patients with Carvajal syndrome suffered from heart failure in their teenage years, resulting in early morbidity. However, further research revealed that it is characterized mainly by ventricular hypertrophy, ventricular dilatation, and discrete focal ventricular aneurysms. In the right ventricle in particular, focal wall thinning and aneurysmal dilatation were identified in the triangle of dysplasia.

5.6.9.3 Left Dominant ARVD/C (LDAC)

As mentioned earlier, the histologic process in classic ARVD/C predominantly involves the RV and extends to the LV in more advanced stages.[52, 59, 66–68] In contrast, patients with left-dominant arrhythmogenic cardiomyopathy (LDAC, also known as left-sided ARVD/C or arrhythmogenic left ventricular cardiomyopathy) have fibrofatty changes that predominantly involve the LV.[68] Clinically, this disease entity is characterized by (infero) lateral T-wave inversion, arrhythmia of LV origin, and/or proven LDAC.

Patients presented with arrhythmia or chest pain at ages ranging from adolescence to over 80 years. By cardiovascular MRI, about one third of patients show an LV ejection fraction <50%. Furthermore, MRI with late gadolinium enhancement (LGE) of the LV demonstrated late enhancement in a subepicardial/midwall distribution. Similar to ARVD/C, some patients with LDAC have desmosomal gene mutations (see later).

5.7 Differential Diagnosis

Although diagnosis in an overt case of ARVD/C is often not difficult, early and occasionally late stages of the disease may show similarities with a few other diseases. Especially differentiation from idiopathic VT originating from the RV outflow tract (RVOT) can be challenging. However, *idiopathic RVOT VT* is a benign nonfamilial condition, in which the ECG shows no depolarization or repolarization abnormalities and no RV structural changes can be detected. Furthermore, VT episodes have a single morphology (LBBB morphology with inferior axis) and are usually not reproducibly inducible by premature extrastimuli at programmed stimulation during EP studies.[70, 71] In contrast, idiopathic RVOT VT may be inducible by regular burst pacing and isoproterenol infusion. It is important to differentiate idiopathic RVOT VT from ARVD/C for several reasons. The first is the known genetic etiology in ARVD/C whereas RVOT tachycardia has not. Therefore, it has implications with regard to screening of family members. The

prognosis of RVOT tachycardia is uniformly excellent with sudden death occurring extremely rarely. Finally, in contrast to ARVD/C, catheter ablation is usually curative in idiopathic RVOT tachycardia.

Another disease mimicking ARVD/C is cardiac *sarcoidosis.* Sarcoidosis is a disease with unknown etiology, characterized by the presence of noncaseating granulomas in affected tissues; mainly lungs, but heart, skin, eyes, reticuloendothelial system, kidneys, and central nervous system can also be affected. The prevalence of this condition varies between geographical regions, and the disease may also be familial and occurring in specific racial subgroups.[72] Clinical symptoms of cardiac involvement are present in about 5% of all patients with sarcoidosis. The clinical manifestations of cardiac sarcoidosis depend on the location and extent of granulomatous inflammation and include conduction abnormalities, ventricular arrhythmias, valvular dysfunction, and congestive heart failure. Myocardial sarcoid granulomas or areas of myocardial scarring are typically present in the left ventricle and septum of patients with this condition, yet the right ventricle can be predominantly affected. A VT associated with right ventricular abnormalities can, therefore, result in diagnostic confusion, especially if there is no systemic evidence of sarcoidosis. Patients can present with clinical features similar to those of ARVD/C including arrhythmias and sudden cardiac death.[73] Cardiac sarcoidosis can only be diagnosed definitively by endomyocardial biopsy, when granulomas are visualized. To strengthen differentiation from ARVD/C, gadolinium-enhanced MRI may be beneficial by detecting located abnormalities in the septum, which is typical for sarcoidosis but hardly ever seen in ARVD/C. Active foci of sarcoidosis can be visualized by positron emission (PET) scan. Therapy with corticosteroids is recommended for patients with a clear diagnosis of cardiac sarcoidosis. Treatment aims to control inflammation and fibrosis in order to maintain cardiac structure and function.

Also, any other form of *myocarditis* has to be excluded before diagnosis of ARVD/C can be made. Myocarditis may arise from viral or other pathogen exposure as well as toxic or immunologic insult. In general, endomyocardial biopsy is required to distinguish it from ARVD/C.

Especially in more advanced stages of the disease, when LV ejection fraction drops below 50%, ARVD/C may mimic dilated cardiomyopathy (DCM). Patients with DCM usually present with heart failure or thromboembolic disease, including stroke. It is uncommon to have sustained ventricular tachycardia or sudden death as the initial presenting symptom of DCM. Therefore, in that case patients should be first suspected to have ARVD/C.

5.8 Molecular Genetic Analysis

It is important to realize that ARVD/C diagnosis is based on the clinical diagnostic TFC. To date, molecular information has not been incorporated into the diagnostic criteria. Mutations underlying the disease show incomplete penetrance and variable clinical expression. Some genetically affected patients can have no signs or symptoms whatsoever, whereas no mutations can be identified in a large minority of clinically diagnosed patients. Therefore, DNA analyses will not be of any consequence for the index patient, but can be used to identify whether family members are predisposed to disease development.

The strategy for genetic testing in ARVD/C is as follows.

Individuals with clinical diagnosis of ARVD/C are the first to be tested. The detection of a pathogenic mutation does not make a diagnosis of ARVD/C. In contrast, if no mutation can be identified in a patient diagnosed with ARVD/C, the clinical diagnosis of ARVD/C is still applicable. In case a pathogenic mutation is identified in the proband, parents, siblings, and possibly children of this patient can be subsequently tested for the mutation concerned, via the cascade method. When an (asymptomatic) relative is found to carry a pathogenic mutation, cardiologic screening is required at least biannually.

Table 5.1 shows the different genes related to ARVD/C. Although numbers vary per country, *PKP2* accounts for the large majority of mutations found. Currently, DNA analysis on *PKP2*, *DSG2*, *DSC2*, *DSP*, and *JUP* is recommended in all ARVD/C patients.

5.9 Prognosis and Therapy

Although the prognosis of ARVD/C is considerably better than that of sustained ventricular tachycardia with left ventricular structural heart disease, ARVD/C

is a progressive disease and will probably lead to right ventricular failure in the long term unless sudden cardiac death occurs. The death rate for patients with ARVD/C has been estimated at 2.5% per year.[74] Retrospective analysis of clinical and pathologic studies identified several risk factors for sudden death, such as previously aborted sudden death, syncope, young age, malignant family history, severe RV dysfunction, and LV involvement.[75,76]

Electrophysiologic testing by programmed ventricular stimulation can be useful for diagnostic purposes by induction of multiple VT morphologies and VT with LBBB morphologies and superior axis.[57] However, EP studies have proven not to be useful in risk stratifying patients with ARVD/C. This was illustrated in the multicenter study of 132 patients with ARVD/C in whom electrophysiologic study was performed prior to ICD implantation.[77] The positive and negative predictive values of VT inducibility for subsequent appropriate device therapy were only 49% and 54%, respectively.

In addition to symptomatic treatment, prevention of sudden death is the most important therapeutic goal in ARVD/C. Most data available on effective treatment strategies refer to retrospective analyses in single centers with only limited number of patients, of which results are difficult to compare due to different patient selections and treatment strategies. There is limited data on long-term outcomes and no controlled randomized trials have been performed. International registries are established, but have not yet reported results on treatment.

Evidence suggests that asymptomatic patients and healthy mutation carriers do not require any prophylactic treatment. They should instead undergo regular cardiologic check-ups including 12-lead ECG, 24 h Holter monitoring, echocardiography, and exercise testing for early identification of unfavorable signs. To all patients diagnosed with or showing multiple signs of ARVD/C as well as all mutation carriers, specific life style advises have to be given, indifferent from which additional therapeutic measures are taken. Sports participation has been shown to increase the risk of sudden death in ARVD/C patients fivefold.[78] Furthermore, excessive mechanical stress, such as during competitive sports activity and training, may aggravate the underlying myocardial lesion and accelerate disease progression. Therefore, patients with ARVD/C have to be advised against practicing highly competitive sports and sports with long endurance, like running marathons.

Therapeutic options in patients with ARVD/C include anti-arrhythmic drugs, catheter ablation, and implantation of cardioverter defibrillators (*ICDs*).

Patients with recurrences of ventricular tachycardia have a favorable outcome when they are treated medically and therefore pharmacologic treatment is the first choice. This concerns not only patients who have presented with sustained VT, but also patients and family members with nonsustained VT or >500 ventricular extrasystoles on 24-h Holter monitoring. Since ventricular arrhythmias and cardiac arrest occur frequently during or after physical exercise or may be triggered by catecholamines, anti-adrenergic β-blockers are recommended. Sotalol is the drug of first choice. Alternatively, other β-receptor blocking agents, amiodarone and flecainide, have all been reported as useful. Efficacy of drug treatment has to be evaluated by serial Holter monitoring and/or exercise testing. This strategy has proven to have better long-term outcome than standard empirical treatment (Fig. 5.7).[79]

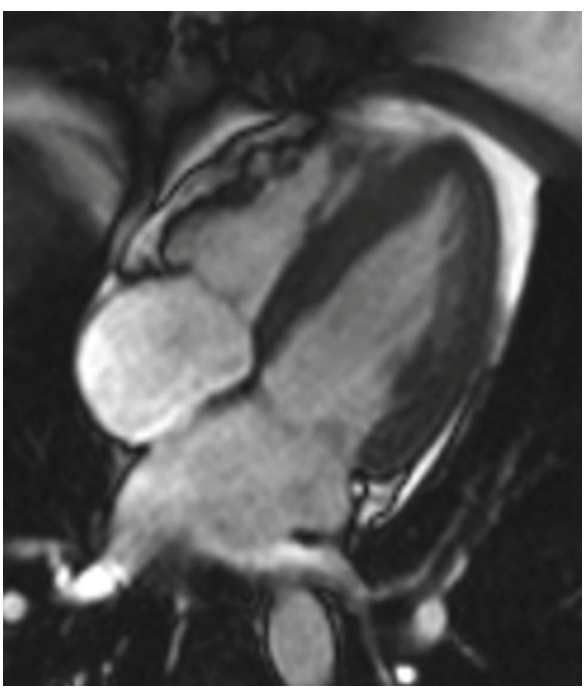

Fig. 5.7 Magnetic resonance imaging (*MRI*) image of arrhythmogenic right ventricular dysplasia/cardiomyopathy (*ARVD/C*) patient at the end of systole. Dyskinetic bulgings are clearly visible in the right ventricle (*RV*) free wall

Catheter ablation is an alternative in patients who are refractory to drug treatment and have frequently recurring VT episodes with predominantly a single morphology. In addition, catheter ablation has been shown to improve the effectiveness of pharmacological treatment: 70% of patients may respond to anti-arrhythmic agents to whom they were unresponsive prior to ablation therapy.[80] Marchlinski et al. performed VT ablation in 19 ARVD/C patients by use of focal and/or linear lesions, in 17 of whom no VT recurred during the subsequent 7 ± 22 months.[81] In a series of 50 consecutive patients studied during 16 years, Fontaine et al. reached a 40% success rate by radiofrequency ablation after multiple ablation sessions, which increased to 81% when fulguration was used additionally.[82] However, these successes have been reported by single centers with highly experienced electrophysiologists, and may not hold true in general practice. Catheter ablation is usually considered as only palliative and not curative. In general, long-term success rates are poor. Owing to disease progression, new VTs with different morphologies will occur after a certain period of time.[83]

Although *anti-arrhythmic drugs* and catheter ablation may reduce VT burden, there is no proof from prospective trials that these therapies will also prevent sudden death. ICD implantation is indicated in patients who are intolerant to anti-arrhythmic drug therapy and who are at serious risk for sudden death. Implantation of an ICD has to be considered in ARVD/C patients with aborted cardiac arrest, intolerable fast VT, and those with risk factors as mentioned above.

5.10 Summary

ARVD/C is most often a genetically determined disease characterized by fibrofatty replacement of myocardial tissue. Clinically, it affects primarily the right ventricle, but extension to the left ventricle occurs, especially in more advanced stages of the disease. Patients typically present between the second and fourth decade of life with exercise-induced tachycardias originating from the right ventricle. However, it is also a major cause of sudden death in the young and athletes.

Its prevalence has been estimated to vary from 1:2,000 to 1:5,000.

The causative genes encode proteins of mechanical cell junctions (plakoglobin, plakophilin2, desmoglein, desmocollin, desmoplakin) and account for intercalated disk remodeling. The classical form of ARVD/C is inherited in an autosomal dominant trait, but shows reduced penetrance and variable expression. The more rare recessively inherited variants are often associated with palmoplantar keratoderma and woolly hair. Clinical diagnosis is made according to a set of Task Force criteria, based on family history, depolarization and repolarization abnormalities, arrhythmias with a left bundle branch block morphology, functional and structural alterations of the right ventricle, and fibrofatty replacement through endomyocardial biopsy. Two-dimensional echocardiography, cine-angiography, and magnetic resonance are the imaging tools for visualizing structural/functional abnormalities. The main differential diagnoses are idiopathic right ventricular outflow tract tachycardia, myocarditis, and sarcoidosis. Only palliative therapy is available and consists of anti-arrhythmic drugs, catheter ablation, and implantable cardioverter defibrillator. Young age, family history of juvenile sudden death, overt left ventricular involvement, ventricular tachycardia, syncope, and previous cardiac arrest are the major risk factors for adverse prognosis.

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