

R E V I E W

Cardiomyopathies

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Summary. The most common cardiomyopathies often present to primary care physicians with similar symptoms, despite the fact that they involve a variety of phenotypes and etiologies (1). Many have signs and symptoms common in heart failure, such as reduced ejection fraction, peripheral edema, fatigue, orthopnea, exertion dyspnea, paroxysmal nocturnal dyspnea, presyncope, syncope and cardiac ischemia (1). In all cardiomyopathies, the cardiac muscle (myocardium) may be structurally and/or functionally impaired. They can be classified as hypertrophic, dilated, left-ventricular non compaction, restrictive and arrhythmogenic right ventricular cardiomyopathies. (www.actabiomedica.it)

Key words: Hypertrophic cardiomyopathy, dilated cardiomyopathy, restrictive cardiomyopathy, left ventricular noncompaction, arrhythmogenic right ventricular cardiomyopathy

Hypertrophic cardiomyopathy

Hypertrophic cardiomyopathy (CMH) is characterized by an increase in the number of heart muscle cells. It is frequently caused by mutations in genes encoding sarcomeric proteins, leading to myocyte disarray, a hallmark of CMH (2).

Clinical symptoms range from asymptomatic left ventricular hypertrophy to progressive heart failure or sudden cardiac death, and vary from individual to individual even within the same family. Frequent symptoms include dyspnea, chest pain, palpitations, orthostasis, presyncope and syncope. Usually CMH becomes apparent during adolescence or early adulthood, although it may also develop in different stages of life such as old age, infancy or childhood (3).

Hypertrophic cardiomyopathy is a relatively common inherited heart disease with a prevalence of 1:500 in the population (4). Clinical diagnosis is based on

patient history, physical examination, echocardiography and ECG to detect hypertrophy (2). The genetic test is useful for confirming diagnosis, and for differential diagnosis, recurrence risk evaluation and prenatal diagnosis in families with a known mutation. Differential diagnosis should consider acquired left ventricular hypertrophy, Danon disease, Fabry disease, cardiac amyloidosis, glycogen storage disease type II, Noonan syndrome and Friedreich ataxia (5).

The European Society of Cardiology recommends genetic testing in the following cases (6):

- 1 - patients meeting diagnostic criteria for CMH, when testing enables cascade genetic screening of their relatives;
- 2 - in first-degree adult relatives of patients with a definite disease-causing variant;
- 3 - in first-degree adult relatives, clinical screening with ECG and echocardiogram should be offered when genetic testing is not performed

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in the proband, or when genetic analysis fails to identify a definite mutation or reveals one or more genetic variants of unknown significance;

- 4 - children of patients with a definite disease-causing mutation should be considered for predictive genetic testing after pre-test family counseling when they are at least 10 years old;
- 5 - when there is a family history of childhood malignancies or early-onset disease or when children have heart symptoms or are involved in particularly demanding physical activity, clinical or genetic testing of first-degree child relatives may be considered before the age of 10 years.

Hypertrophic cardiomyopathy typically has autosomal dominant inheritance. Pathogenic variants may be missense, nonsense, splicing or small indels (Table 1). Large deletions/duplications have also been reported in the *NEXN*, *TNNI3*, *MYBPC3*, *CAV3* and *MYH7* genes.

The mutation detection rate for the most common mutant genes is ~56% (*MYBPC3* 20-30%; *MYH7* 20-30%; *TNNT2* 3-5%; *TNNI3* 3-5%; *TPM1* 1-3%) (7). MAGI uses a multi-gene NGS panel to detect nucleotide variations in coding exons and flanking introns of the above genes, and MLPA to detect duplications and deletions in *NEXN*, *TNNI3*, *MYBPC3*, *CAV3* and *MYH7*. Worldwide, 151 accredited medical genetic laboratories in the EU and 19 in the US, listed in the Orphanet (8) and GTR (9) databases, respectively, offer genetic tests for hypertrophic cardiomyopathy. The clinical guidelines for genetic testing are described in Genetics Home Reference (10), GeneReviews (5) and Clinical Utility Gene Card (7).

Dilated cardiomyopathy

Dilated cardiomyopathy (CMD) is a heart disorder characterized by dilation of at least one ventricle and systolic dysfunction. The ventricle wall becomes thinner and its contractile force decreases. Clinical signs are usually arrhythmias, thromboembolic events, such as stroke, and above all symptoms of heart failure, such as edema, orthopnea, dyspnea and fatigue. How-

ever, the symptoms take years to cause health problems and severity varies between affected individuals.

The etiology of CMD may include either inherited or acquired causes, such as myocardial infarction, valve disease, toxins, drugs, inflammatory conditions, long-standing severe hypertension and irradiation of the chest (11). Dilated cardiomyopathy is essentially an adult-onset disease, but has shown a highly variable age of onset (12). The prevalence is 1:2700 (13). It can be classified as acquired, syndromic or non syndromic.

Diagnosis is established when left ventricular enlargement and systolic dysfunction are both ascertained. Patient history, physical examination and echocardiography are also indispensable for the diagnostic process (12). The genetic test is useful for diagnosis confirmation, differential diagnosis, recurrence risk evaluation and prenatal diagnosis in families with a known mutation. Differential diagnosis should consider acquired dilated cardiomyopathies, syndromic forms and other cardiomyopathies that may present with left ventricular involvement (14). Syndromic forms include *HFE*-associated hereditary hemochromatosis, Emery-Dreifuss muscular dystrophy, Laing distal myopathy, Carvajal syndrome, Duchenne and Becker muscular dystrophy, Barth syndrome and mitochondrial dilated cardiomyopathies (15).

Dilated cardiomyopathy is a genetically heterogeneous disease and has different modes of inheritance (Table 2). Pathogenic variants may be missense, nonsense, splicing and small indels. Large deletions/duplications have also been reported in *LMNA*, *MYH7*, *SCN5A*, *BAG3*, *DES*, *EYA4*, *SGCD*, *MYBPC3*, *NEXN*, *PRDM16*, *PSEN1*, *TNNI3*, *DND*, *RAF1*, *FKTN* and *TAZ*. The mutation detection rates for the most frequently mutant CMD-related genes are *TTN* 18-25%, *LMNA* 6%, *MYH7* 4-5%, *MYH6* 3-4%, *MYBPC3* 2-4%, *TNNT2* 3%, *BAG3* 2-3%. (16).

Our multi-gene NGS panel to detect nucleotide variations in coding exons and flanking introns of the above genes, MLPA to detect duplications and deletions in *LMNA*, *MYH7*, *SCN5A*, *BAG3*, *DES*, *EYA4*, *SGCD*, *MYBPC3*, *NEXN*, *PRDM16*, *PSEN1*, *TNNI3*, *DND*, *RAF1*, *FKTN* and *TAZ*.

Worldwide, 49 accredited medical genetic laboratories in the EU and 44 in the US, listed in the Orphanet (8) and GTR (9) databases, respectively, offer

Table 1. Genes associated with various forms of hypertrophic cardiomyopathy

| Gene | OMIM gene | Disease | OMIM disease | Inheritance | Function |
|---------------|-----------|---------|--------------|-------------|--|
| <i>MYH7</i> | 160760 | CMH1 | 192600 | AD | Beta heavy chain subunit of cardiac myosin |
| <i>CAV3</i> | 601253 | CMH1 | 192600 | AD | Regulates voltage-gated K ⁺ channels and plays a role in sarcolemma repair in cardiomyocytes after mechanical stress |
| <i>MYLK2</i> | 606566 | CMH1 | 192600 | AD | Cardiac Ca ²⁺ /calmodulin-dependent myosin light chain |
| <i>TNNT2</i> | 191045 | CMH2 | 115195 | AD | Ca ²⁺ -dependent regulator of muscle contraction |
| <i>TPM1</i> | 191010 | CMH3 | 115196 | AD | Ca ²⁺ -dependent regulator of striated muscle contraction |
| <i>MYBPC3</i> | 600958 | CMH4 | 115197 | AD | Cardiac isoform of myosin-binding protein C found in cross-bridge-bearing zone (C region) of A bands |
| <i>PRKAG2</i> | 602743 | CMH6 | 600858 | AD | Energy-sensing enzyme that monitors cell energy status and functions. Inhibitor of de novo biosynthesis of fatty acids and cholesterol |
| <i>TNNI3</i> | 191044 | CMH7 | 613690 | AD | Cardiac mediator of striated muscle relaxation |
| <i>MYL3</i> | 160790 | CMH8 | 608751 | AD | Ventricular isoform of myosin light chain 3 |
| <i>TTN</i> | 188840 | CMH9 | 613765 | AD | Important for assembly and functioning of striated muscles, it connects microfilaments and contributes to balance of forces between two halves of sarcomere |
| <i>MYL2</i> | 160781 | CMH10 | 608758 | AD | Regulatory light chain associated with cardiac myosin beta heavy chain, promoting cardiac myofibril assembly |
| <i>ACTC1</i> | 102540 | CMH11 | 612098 | AD | ACTC1 is localized in contractile apparatus of muscle tissues |
| <i>CSRP3</i> | 600824 | CMH12 | 612124 | AD | Positive regulator of myogenesis; transcription cofactor for myogenic bHLH transcription factors |
| <i>TNNC1</i> | 191040 | CMH13 | 613243 | AD | TNNC1 encodes Tn-C that abolishes inhibitory action of Tn on actin filaments upon Ca ²⁺ binding |
| <i>MYH6</i> | 160710 | CMH14 | 613251 | AD | Alpha heavy chain subunit of cardiac myosin |
| <i>VCL</i> | 193065 | CMH15 | 613255 | AD | VCL encodes an actin filament-binding protein that regulates cell-matrix adhesion, cell-cell adhesion, cell-surface E-cadherin expression, mechanosensing by E-cadherin complex, cell morphology and cell locomotion |
| <i>MYOZ2</i> | 605602 | CMH16 | 613838 | AD | MYOZ2 encodes myozenin that binds proteins involved in linking Z line proteins and localizing calcineurin signaling to sarcomeres. May play a role in myofibrillogenesis |

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Table 1 (continued). Genes associated with various forms of hypertrophic cardiomyopathy

| Gene | OMIM gene | Disease | OMIM disease | Inheritance | Function |
|--------------|-----------|----------------------------|--------------|-------------|--|
| <i>JPH2</i> | 605267 | CMH17 | 613873 | AD | JPH2 is necessary for intracellular Ca ²⁺ signaling in cardiac myocytes via ryanodine receptor-mediated Ca ²⁺ release |
| <i>PLN</i> | 172405 | CMH18 | 613874 | AD | Modulates contractility of heart muscle in response to physiological stimuli via ATP2A2 regulates Ca ²⁺ re-uptake during muscle relaxation and Ca ²⁺ homeostasis in heart muscle |
| <i>CALR3</i> | 611414 | CMH19 (?) | 613875 | AD | Ca ²⁺ -binding chaperone localized in endoplasmic reticulum |
| <i>NEXN</i> | 613121 | CMH20 | 613876 | AD | Essential for maintenance of sarcomere integrity |
| <i>MYPN</i> | 608517 | CMH22 | 615248 | AD | Component of cardiac muscle sarcomere that links nebullette to alpha-actinin in Z lines |
| <i>ACTN2</i> | 102573 | CMH23 with or without LVNC | 612158 | AD | Localized in Z-disc of cardiac muscle where it anchors myofibrillar actin filaments |
| <i>LDB3</i> | 605906 | CMH24 | 601493 | AD | Adaptor protein in striated muscle; couples protein kinase C-mediated signaling to cytoskeleton |
| <i>TCAP</i> | 604488 | CMH25 | 607487 | AD | Muscle assembly regulating factor that mediates antiparallel assembly of titin molecules at sarcomere Z-disk |
| <i>FLNC</i> | 102565 | CMH26 | 617047 | AD | Critical for myogenesis and structural integrity of muscle fibers |

CMH=hypertrophic cardiomyopathy; LVNC=left ventricular non-compaction; AD=autosomal dominant; AR=autosomal recessive

genetic testing for CMD. The guidelines for clinical use of genetic testing are described in Genetics Home Reference (10), GeneReviews (12) and Clinical Utility Gene Card (16).

Restrictive cardiomyopathy

Restrictive cardiomyopathy (RCM) is a rare genetic heart disease characterized by restrictive ventricle filling and diastolic dysfunction due to cardiac muscle stiffness which leads to abnormal relaxation of the ventricles, although thicknesses and systolic func-

tion are usually normal until later stages of the disease (17). It can manifest at any time from childhood to adulthood. In children, the first signs may be failure to gain weight and thrive, fatigue and fainting. As the disease advances, there may be edema, ascites, hepatomegaly and lung congestion. Some children are totally asymptomatic and sudden death is the first manifestation. Adults with RCM first develop dyspnea, fatigue and reduced ability to exercise. Arrhythmia and palpitations are also typical of adults with RCM (18). Restrictive cardiomyopathy is uncommon: in the US and Europe, it accounts for less than 5% of all cardiomyopathies. Prevalence is unknown (19).

Table 2. Genes associated with various forms of dilated cardiomyopathies

| Gene | OMIM gene | Disease | OMIM disease | Inheritance | Function |
|--------------|-----------|--------------------------|--------------|-------------|---|
| <i>LMNA</i> | 150330 | CMD1A | 115200 | AD | Required for cardiac homeostasis |
| <i>MYH7</i> | 160760 | CMD1S | 613426 | AD | Beta heavy chain subunit of cardiac myosin |
| <i>MYH6</i> | 160710 | CMD1EE | 613252 | AD | Alpha heavy chain subunit of cardiac myosin. |
| <i>SCN5A</i> | 600163 | CMD1E | 601154 | AD | Mediates voltage-dependent Na ⁺ permeability of excitable membranes |
| <i>ACTN2</i> | 102573 | CMD1AA with/without LVNC | 612158 | AD | Localized in the Z-disc of cardiac muscle where it anchors myofibrillar actin filaments |
| <i>DSG2</i> | 125671 | CMD1BB | 612877 | AD | Ca ²⁺ -binding transmembrane glycoprotein component of desmosomes between myocardial cells |
| <i>LDB3</i> | 605906 | CMD1C with/without LVNC | 601493 | AD | Adaptor protein in striated muscle; couples protein kinase C-mediated signaling to cytoskeleton |
| <i>TNNT2</i> | 191045 | CMD1D | 601494 | AD | Ca ²⁺ -dependent regulator of muscle contraction |
| <i>RBM20</i> | 613171 | CMD1DD | 613172 | AD | RNA-binding protein that regulates mRNA splicing of genes involved in heart development, such as TTN |
| <i>TTN</i> | 188840 | CMD1G | 604145 | AD | Important for striated muscle assembly and function, connects microfilaments, contributes to balance of forces between two halves of sarcomere |
| <i>BAG3</i> | 603883 | CMD1HH | 613881 | AD | Co-chaperone for HSP70 and HSC70 chaperone proteins in heart; triggers client/substrate protein release |
| <i>DES</i> | 125660 | CMD1I | 604765 | AD | Sarcomeric microtubule-anchoring protein that maintains sarcomere structure |
| <i>CRYAB</i> | 123590 | CMD1II | 615184 | AD | Has chaperone-like activity, preventing aggregation of proteins under stress conditions |
| <i>EYA4</i> | 603550 | CMD1J | 605362 | AD | Transcriptional regulator during organogenesis |
| <i>LAMA4</i> | 600133 | CMD1JJ | 615235 | AD | Mediates attachment, migration and organization of cells into tissues during embryo development by interacting with other extracellular matrix components |
| <i>MYPN</i> | 608517 | CMD1KK | 615248 | AD | Component of heart muscle sarcomere linking nebullette to alpha-actinin in Z lines |
| <i>SGCD</i> | 601411 | CMD1L | 606685 | AD | Component of sarcoglycan complex linking F-actin cytoskeleton and extracellular matrix |
| <i>CSRP3</i> | 600824 | CMD1M | 607482 | AD | Positive regulator of myogenesis; transcription cofactor for myogenic bHLH transcription factors |
| <i>ABCC9</i> | 601439 | CMD1O | 608569 | AD | Activates and regulates cardiac and smooth muscle-type KATP channels |

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Table 2 (continued). Genes associated with various forms of dilated cardiomyopathies

| Gene | OMIM gene | Disease | OMIM disease | Inheritance | Function |
|---------------|-----------|---|----------------|-------------|--|
| <i>PLN</i> | 172405 | CMD1P | 609909 | AD | Modulates contractility of heart muscle in response to physiological stimuli via ATP2A2; regulates Ca ²⁺ re-uptake during muscle relaxation and Ca ²⁺ homeostasis in heart muscle |
| <i>ACTC1</i> | 102540 | CMD1R | 613424 | AD | Localized in contractile apparatus of muscle tissue |
| <i>MYBPC3</i> | 600958 | CMD1MM | 615396 | AD | Cardiac isoform of myosin-binding protein C found in cross-bridge-bearing zone (C region) of A bands |
| <i>PRDM16</i> | 605557 | CMD1LL | 615373 | AD | Transcriptional cofactor essential for heart development |
| <i>PSEN1</i> | 104311 | CMD1U | 613694 | AD | Expressed in heart and critical for heart development |
| <i>PSEN2</i> | 600759 | CMD1V | 613697 | AD | Expressed in heart and critical for heart development |
| <i>TPM1</i> | 191010 | CMD1Y | 611878 | AD | Ca ²⁺ -dependent regulator of striated muscle contraction |
| <i>VCL</i> | 193065 | CMD1W | 611407 | AD | Encodes an actin filament-binding protein that regulates cell-matrix adhesion, cell-cell adhesion, cell-surface E-cadherin expression, mechanosensing by the E-cadherin complex, cell morphology and cell locomotion |
| <i>TNNC1</i> | 191040 | CMD1Z | 611879 | AD | Encodes Tn-C that abolishes inhibitory action of Tn on actin filaments upon Ca ²⁺ binding |
| <i>RAF1</i> | 164760 | CMD1NN | 615916 | AD | Promotes cardiomyocyte survival |
| <i>DSP</i> | 125647 | CMD with woolly hair, keratoderma, tooth agenesis | 615821, 605676 | AD, AR | Obligate component of functional desmosomes |
| <i>TCAP</i> | 604488 | CMD | / | AD | Muscle assembly regulating factor that mediates antiparallel assembly of titin molecules at sarcomeric Z-disk |
| <i>ANKRD1</i> | 609599 | CMD | / | AD | Nuclear negative transcription factor that regulates expression of cardiac genes |
| <i>TMPO</i> | 188380 | CMD | / | AD | Regulates expression patterns of major cardiac transcription factors |
| <i>ILK</i> | 602366 | CMD | / | AD | Migration and survival of myocardial and endothelial cells |
| <i>TNNI3</i> | 191044 | CMD2A, CMD1FF | 611880, 613286 | AR | Cardiac mediator of striated muscle relaxation |

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Table 2 (continued). Genes associated with various forms of dilated cardiomyopathies

| Gene | OMIM gene | Disease | OMIM disease | Inheritance | Function |
|---------------|-----------|---------|--------------|-------------|---|
| <i>GATAD1</i> | 614518 | CMD2B | 614672 | AR | Regulates gene expression by binding to a histone modification site |
| <i>FKTN</i> | 607440 | CMD1X | 611615 | AR | Glycosylation of alpha-dystroglycan in skeletal muscle |
| <i>SDHA</i> | 600857 | CMD1GG | 613642 | AR | Major catalytic subunit of succinate-ubiquinone oxidoreductase located in mitochondrial respiratory chain |
| <i>DMD</i> | 300377 | CMD3B | 302045 | XLR | Anchors extracellular matrix to cytoskeleton via F-actin |
| <i>TAZ</i> | 300394 | CMD | / | XLR | Involved in cardiolipin metabolism |

CMD=dilated cardiomyopathy, LVNC=left ventricular non-compaction AD=autosomal dominant; AR=autosomal recessive; XLR=X-linked recessive.

Clinical diagnosis is based on medical and family history, physical examination, chest X-ray, echocardiography, ECG, Holter monitoring, stress test, cardiac MRI, cardiac catheterization, coronary angiography and myocardial biopsy (18). Genetic testing is useful for confirming diagnosis, and for differential diagnosis, recurrence risk evaluation and prenatal diagnosis in families with a known mutation. Differential diagnosis should consider constrictive pericarditis, idiopathic forms, such as Loeffler eosinophilic endomyocardial disease, secondary forms, such as infiltrative disease (amyloidosis, sarcoidosis, hemochromatosis, Fabry disease, Danon disease and Friedreich ataxia) and treatment-induced RCM (post-irradiation fibrosis and drug-induced RCM) (19).

Restrictive cardiomyopathy typically has autosomal dominant inheritance (Table 3). Pathogenic variants may be missense, nonsense, splicing and small indels. Large deletions/duplications have been reported in *TNNI3*, *MYBPC3* and *MYH7*. MAGI uses a multi-gene NGS panel to detect nucleotide variations in coding exons and flanking introns of the above genes, and MLPA to detect duplications and deletions in the *TNNI3*, *MYBPC3* and *MYH7* genes. 6 accredited medical genetic laboratories in the US, listed in the GTR (9) database, offer genetic tests for RCM.

The guidelines for clinical use of genetic testing are described in Genetics Home Reference (10).

Left ventricular noncompaction

Left ventricular noncompaction (LVNC) is a heart disorder that affects the cardiac muscle, mostly the left ventricle, which acquires a thick spongy appearance. The disease is considered to be a consequence of an arrest in heart development during embryogenesis (20). The abnormal cardiac muscle does not function properly, leading to progressive systolic and diastolic dysfunction. LVNC may be isolated or an element of other heart diseases.

The disorder has a variety of symptoms. Some patients may be entirely asymptomatic, while others fall victim to sudden death. Other symptoms or signs may be arrhythmia, palpitations, abnormal blood clots, fatigue, dyspnea and lymphedema (21). Although the disease is genetic, age of onset is variable and diagnosis may be made from birth to late adulthood. The prevalence of LVNC is less than 0.25% (22).

Clinical diagnosis is mainly based on structural features observed by cardiac imaging. Echocardiography is used for diagnosis and follow-up. MRI can

Table 3. Genes associated with various forms of restrictive cardiomyopathy

| Gene | OMIM gene | Disease | OMIM disease | Inheritance | Function |
|---------------|-----------|---------|--------------|-------------|--|
| <i>TNNI3</i> | 191044 | RCM1 | 115210 | AD | Cardiac mediator of striated muscle relaxation |
| <i>TNNT2</i> | 191045 | RCM3 | 612422 | AD | Ca ²⁺ -dependent regulator of muscle contraction |
| <i>MYPN</i> | 608517 | RCM4 | 615248 | AD | Component of the heart muscle sarcomere linking nebulin to alpha-actinin in Z lines |
| <i>FLNC</i> | 102565 | RCM5 | 617047 | AD | Critical for myogenesis and structural integrity of muscle fibers |
| <i>ACTC1</i> | 102540 | RCM | / | AD | Localized in contractile apparatus of muscle tissue |
| <i>MYH7</i> | 160760 | RCM | / | AD | Beta heavy chain subunit of cardiac myosin |
| <i>MYBPC3</i> | 600958 | RCM | / | AD | Cardiac isoform of myosin-binding protein C found in cross-bridge-bearing zone (C region) of A bands |
| <i>TPM1</i> | 191010 | RCM | / | AD | Ca ²⁺ -dependent regulator of striated muscle contraction |
| <i>MYL1</i> | 160780 | RCM | / | AD | Regulatory light chain of myosin |
| <i>MYL2</i> | 160781 | RCM | / | AD | Regulatory light chain associated with cardiac myosin beta heavy chain, promoting cardiac myofibril assembly |

RCM=restrictive cardiomyopathy; AD=Autosomal dominant.

be useful in cases with poor echocardiogram findings. Genetic testing is useful for confirming diagnosis and for differential diagnosis, recurrence risk evaluation and prenatal diagnosis in families with a known mutation. Differential diagnosis should consider dilated cardiomyopathy, hypertensive heart disease, apical hypertrophic cardiomyopathy, infiltrative cardiomyopathy, eosinophilic endomyocardial disease, localized left ventricular hypertrophy, left ventricular thrombi, cardiac metastases, endocardial fibroelastosis and Barth syndrome (23).

Left ventricular noncompaction is a genetically heterogeneous disorder with sporadic and familial forms (24). Autosomal dominant inheritance seems more common than X-linked inheritance (25). Autosomal recessive inheritance and mitochondrial inheritance have also been observed (26). Current evidence suggests that in most cases, an association with genetic cardiomyopathy (CMP) and/or congenital heart dis-

ease (CHD) is more likely than a causal role. Consequently, the genetic basis coincides or overlaps with those of CMP or CHD (27). LVNC has mostly autosomal dominant inheritance, but may also have autosomal recessive inheritance (Table 4).

Pathogenic variants may be sequence variations (missense, nonsense, splicing, small insertions and deletions, small indels). Large deletions/duplications have also been reported in *MYBPC3*, *MYH7*, *PKP2* and *PRDM16*. MAGI uses a multi-gene NGS panel to detect nucleotide variations in coding exons and flanking introns of the above genes, and MLPA to detect duplications and deletions in the same genes.

Worldwide, 40 accredited medical genetic laboratories in the EU and 4 in the US, listed in the Orphanet (8) and GTR (9) databases, respectively, offer genetic testing for LVNC. The guidelines for clinical use of genetic testing are described in Genetics Home Reference (10).

Table 4. Genes associated with various forms of left ventricular noncompaction

| Gene | OMIM gene | Disease | OMIM disease | Inheritance | Function |
|----------------|-----------|---------|--------------|-------------|--|
| <i>MYH7</i> | 160760 | LVNC5 | 613426 | AD | Beta heavy chain subunit of cardiac myosin |
| <i>MYBPC3</i> | 600958 | LVNC10 | 615396 | AD | Cardiac isoform of myosin-binding protein C found in cross-bridge-bearing zone (C region) of A bands |
| <i>TPM1</i> | 191010 | LVNC9 | 611878 | AD | Ca ²⁺ -dependent regulation of striated muscle contraction |
| <i>PRDM16</i> | 605557 | LVNC8 | 615373 | AD | Transcriptional cofactor essential for heart development |
| <i>MIB1</i> | 608677 | LVNC7 | 615092 | AD | Involved in heart looping process |
| <i>TNNT2</i> | 191045 | LVNC6 | 601494 | AD | Ca ²⁺ -dependent regulator of muscle contraction |
| <i>ACTC1</i> | 102540 | LVNC4 | 613424 | AD | Localized in muscle tissue contractile system |
| <i>LDB3</i> | 605906 | LVNC3 | 601493 | AD | Adapter protein in striated muscle; couples protein kinase C-mediated signaling to cytoskeleton |
| <i>DTNA</i> | 601239 | LVNC1 | 604169 | AD | Component of dystrophin-associated protein complex; localized in sarcolemma |
| <i>LMNA</i> | 150330 | LVNC | / | AD | Required for cardiac homeostasis |
| <i>SCN5A</i> | 600163 | LVNC | / | AD | Mediates voltage-dependent Na ⁺ permeability of excitable membranes |
| <i>HCN4</i> | 605206 | LVNC | / | AD | Necessary for heart pacemaking |
| <i>PLEKHM2</i> | 609613 | LVNC | / | AR | Regulates conventional kinesin activity |
| <i>PKP2</i> | 602861 | LVNC | / | AR | Plays a role in junctional plaques |
| <i>SOX6</i> | 607257 | LVNC | / | AR | Transcriptional activator required for maintenance of cardiac muscle cells |
| <i>MT-ND1</i> | 516000 | LVNC | / | MT | Core subunit of mitochondrial membrane respiratory chain NADH dehydrogenase |

LVNC=left ventricular noncompaction; AD=Autosomal dominant; AR=Autosomal recessive; MT=Mitochondrial.

Arrhythmogenic right ventricular cardiomyopathy

Arrhythmogenic right ventricular cardiomyopathy (ARVC) is a genetic heart disease characterized by replacement of right ventricular myocytes with fibrous and fatty tissue. This predisposes young persons and athletes to ventricular tachycardia and even sudden death. Symptoms are not frequent in the early

stages, but there is nevertheless risk of sudden death during intense exercise. When symptoms occur, they often include palpitations and syncope. Shortness of breath, swelling of the legs or heart failure are typical of a later stage of the disease. Patients usually develop symptoms between the second and fifth decade. The mean age at diagnosis is 31 years (28).

Prevalence of ARVC is estimated at 1:1000-1250

in the general population (29), but in countries with intensive family screening this disease appears to be much more common (30). Study of a population in which males and females were equally distributed revealed that males were 3.3-fold more likely to be associated with episodes of arrhythmia (31). Expression of the disease is variable, while penetrance is incomplete and age-related (32).

To establish diagnosis, an International Task Force proposed criteria for clinical diagnosis of ARVC/dysplasia that facilitated recognition and interpretation of its often nonspecific clinical features. Structural, histological, electrocardiographic, arrhythmic and familial

features of the disease were incorporated into the criteria, divided into major and minor categories according to the specificity of their association with ARVC/dysplasia. This provided a standard on which to base clinical research and genetic studies (33). Differential diagnosis should consider idiopathic right ventricular outflow-tract tachycardia, cardiac sarcoidosis and congenital heart disease leading to right ventricular volume overload (34).

Arrhythmogenic right ventricular cardiomyopathy has mostly autosomal dominant inheritance and only rarely autosomal recessive or digenic inheritance (28). Pathogenic variants in the genes listed in Table 5

Table 5. Genes associated with various forms of arrhythmogenic right ventricular cardiomyopathy

| Gene | OMIM gene | Disease | OMIM disease | Inheritance | Function |
|---------------|-----------|---------|--------------|-------------|--|
| <i>TGFB3</i> | 190230 | ARVC1 | 107970 | AD | Involved in embryogenesis, differentiation, wound healing |
| <i>RYR2</i> | 180902 | ARVC2 | 600996 | AD | Ca ²⁺ channel that releases Ca ²⁺ from sarcoplasmic reticulum into cytoplasm and triggers cardiac muscle contraction |
| <i>TMEM43</i> | 612048 | ARVC5 | 604400 | AD | Maintains nuclear envelope structure |
| <i>DSP</i> | 125647 | ARVC8 | 607450 | AD | Forms obligate component of functional desmosomes |
| <i>PKP2</i> | 602861 | ARVC9 | 609040 | AD | Plays role in junctional plaques |
| <i>DSG2</i> | 125671 | ARVC10 | 610193 | AD | Ca ²⁺ -binding transmembrane glycoprotein components of desmosomes between myocardial cells |
| <i>JUP</i> | 173325 | ARVC12 | 611528 | AD | Common constituent of desmosomes and intermediate junctions |
| <i>CTNNA3</i> | 607667 | ARVC13 | 615616 | AD | Involved in formation of cell-cell adhesion complexes in muscle cells |
| <i>TTN</i> | 188840 | ARVC | / | AD | Important for striated muscle assembly and functioning; connects microfilaments and contributes to balance of forces between two halves of sarcomere |
| <i>DES</i> | 125660 | ARVC | / | AD | Sarcomeric microtubule-anchoring protein that maintains sarcomere structure |
| <i>LMNA</i> | 150330 | ARVC | / | AD | Required for cardiac homeostasis |
| <i>DSC2</i> | 125645 | ARVC11 | 610476 | AD, AR | Major components of desmosomes (cell-cell junctions found in mechanically-stressed cells) |

ARVC=arrhythmogenic right ventricular cardiomyopathy; AD=Autosomal dominant; AR=Autosomal recessive.

have autosomal dominant inheritance (35). Pathogenic variants may be missense, nonsense, splicing, small indels and gross deletions or duplications. MAGI uses a multi-gene NGS panel to detect nucleotide variations in coding exons and flanking introns of the above genes, and MLPA to detect duplications and deletions in *DSP* and *PKP2*.

Worldwide, 46 accredited medical genetic laboratories in the EU and 22 in the US, listed in the Orphanet (8) and GTR (9) databases, respectively, offer genetic testing for ARVC. The guidelines for clinical use of genetic testing are described in Genetics Home Reference (10) and Clinical Utility Gene Card (35).

Conclusions

We created a NGS panel to detect nucleotide variations in coding exons and flanking regions of all the genes associated with cardiac disorders. When a suspect of cardiomyopathy is present, we perform the analysis of all the genes present in this short article.

In order to have a high diagnostic yield, we developed a NGS test that reaches an analytical sensitivity (proportion of true positives) and an analytical specificity (proportion of true negatives) of $\geq 99\%$ (coverage depth $\geq 10x$).

Conflict of interest: Each author declares that he or she has no commercial associations (e.g. consultancies, stock ownership, equity interest, patent/licensing arrangement etc.) that might pose a conflict of interest in connection with the submitted article

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