Hypertrophic cardiomyopathy — repealing tenets in South Africa

JOHANNA C. MOOLMAN-SMOOK, WILLEM J. DE LANGE, PAUL A. BRINK, VALERIE A. CORFIELD

Summary

Hypertrophic cardiomyopathy (HCM), a common primary cardiac disorder with an increased risk of sudden death, affects all population groups in South Africa. Distinct causal mutations in multiple sarcomeric protein-encoding genes correlate with the risk of sudden death. Such genotype/phenotype correlations cannot be extrapolated geographically or ethnically, necessitating the generation of South African-specific data.

We used DNA-based techniques to search for the causal mutations in a panel of South African HCM-affected subjects (37 with unequivocal HCM, 47 with HCM-like disease). Mutations detected were traced in family members and carriers assessed by echocardiography and electrocardiography.

Nine different HCM-causing mutations (5 unique to South Africa, 3 showing a founder effect) were identified in 3 genes in 24 index cases (57% HCM group, 6% HCM-like group). The different mutations were associated with variable hypertrophy, independent of the risk of sudden death. The disease was generally familial and many at-risk mutation carriers did not meet clinical diagnostic criteria for HCM.

Rigorous diagnosis of index cases facilitates detection of causal mutations, which allows for unequivocal DNA-based diagnosis of at-risk family members, regardless of age or clinical status. This permits focused patient management, informed prognostication and realistic counselling for this insidious disease, as well as time and cost savings.

Hypertrophic cardiomyopathy (HCM), a primary cardiac disorder with a genetic basis and an increased risk of sudden death, affects people of all population groups in South Africa. Although its prevalence in this country has not been established, prospective echocardiographic studies in the USA have estimated its prevalence at 1 in 500 in the general population. This makes HCM one of the most common inherited cardiac disorders, and dispels the myth that it is a rare disease.

Historically, HCM has been described as a condition associated with idiopathic myocardial hypertrophy, which primarily affects the left ventricle and which occurs in the absence of other inciting factors, such as hypertension, which could create a similar clinical picture. This feature has been used as the mainstay of clinical diagnosis. In recent years, the application of molecular genetics to the study of HCM has shown that defects (mutations) in any of at least 7 genes are the underlying cause of this disease. These 7
genes all encode proteins that constitute the basic unit of contractility in striated muscle, viz. the sarcomere. Mutations have so far been found in the genes encoding the cardiac beta myosin heavy chain (MYL7), both the regulatory and essential myosin light chains (MYL2 and MYL3), cardiac myosin-binding protein C (MYBPC3), the cardiac troponins T and I (TNNT2 and TNN13) and alpha tropomyosin (TPM). In addition, recent reports have implicated mutations in 2 other sarcomeric protein-encoding genes, namely cardiac alpha-actin (ACTC) and titin (TTN), in the development of HCM. This genetic knowledge has changed the aetiological status of HCM; it is no longer viewed as an idiopathic disease but is described, in molecular terms, as 'a disease of the sarcomere'.

Defining the molecular basis of HCM-causative lesions has heralded the advent of deoxyribonucleic acid (DNA)-based or molecular diagnosis. This in tum has led to comparisons of the correlation of the presence of disease-causing mutations with the expression of clinical features (genotype/phenotype studies), which have resulted in yet more insights into the disease. It has become clear that meeting the hallmark criteria for diagnosis, namely hypertrophy in the absence of other predisposing factors, does not positively correlate with prognosis. More importantly, the absence of a clinical diagnosis does not indicate freedom from risk of sudden death.

Further conceptual changes concerning the nature of the disease have resulted from genotype/phenotype studies 'prospectively performed' in a family setting. Firstly, it has been demonstrated that in most cases the disease is familial, and that relatives of any index patient should be investigated too, as they may be at risk of the clinical consequences of HCM. It is also now clear that family members carrying the same disease-causing mutation demonstrate a broad range of echo- and electrocardiographic features, as well as symptoms. However, certain characteristics do seem to segregate with particular mutations and/or causative genes. Specifically, survival curves show that some mutations are associated with early and frequent cases of sudden death and others with near-normal survival. This may offer an explanation for the earlier observation that some families display a higher frequency of sudden death than others. It now seems clear that mutations in MYH7 are more often associated with echocardiographically detectable overt hypertrophy, but with a frequency of sudden death that varies with the specific mutation. In contrast, mutations in TNNT2 are most associated with subtle to non-detectable hypertrophy, but with a high frequency of early sudden death. Mutations in MYBPC3, on the other hand, are associated with onset of the disease usually after the fourth decade of life. Thereafter, hypertrophy characteristically increases progressively with age, and may result in death from congestive heart failure later in life. As yet, it has not been possible to clinically subgroup the disease profile associated with mutations in other HCM-causing genes.

Consequently, familial HCM is no longer viewed as a clinically homogeneous condition, as it can be stratified into different categories of disease, dependent upon the particular causative lesion. This knowledge has important implications for patient and family management and counselling, and in assessment of the risk of sudden death.

However, despite the correlations that can be drawn between particular genes or mutations and their clinical consequences, several studies have emphasised the dangers of extrapolating data obtained from one geographical region or subpopulation group to another. There is evidence that the same mutation may have a different clinical outcome when present in diverse ethnic groups, due to the influence of environmental and genetic modifiers. A further caution in interpreting genotype/phenotype studies is that in some cases they are based on data collected from a limited sample size. It is therefore considered prudent, whenever possible, to confirm observed genotype/phenotype correlations in a number of large families of the relevant population group before using data as the basis of clinical management and prognostication programmes.

Currently, more than 100 different HCM-causing mutations have been identified in the nine disease-associated genes described. This extreme genetic and allelic heterogeneity is probably the result of both the rate of spontaneous mutations and the frequency of sudden death associated with many HCM-causing mutations that, by conferring a reproductive disadvantage, decrease the existence of numerous families sharing a common ancestral or founder mutation. Most HCM-causing mutations, therefore, have arisen as independent events and are described as private mutations. As a consequence of this, a unique spectrum of mutations is often found within different subpopulation groups; however, genotype/phenotype studies are frequently only available for mutations extant in Europe and North America. The existence of numerous mutations and specific ethnic profiles also complicates the initial DNA-based screening programme that must be followed to detect the disease-associated mutation in any new HCM index case.

Interestingly, in South Africa, where due to earlier population dynamics, founder effects in other diseases are common, three separate cohorts of HCM-affected families, each carrying a different HCM-causing mutation inherited from one of three common ancestors, have been identified. In addition to these founder mutations, at least 6 other private HCM-causative mutations have been detected in this country. In total, 4 of the HCM-causing mutations detected in South Africa have not been reported elsewhere in the world, underscoring the notion of population-specific mutational profiles.

In view of emerging insights concerning HCM and the unique mutational profile existing in South Africa, it is clear that the full spectrum of HCM-causing mutations and associated genotype/phenotype correlations specific to this country must be established. The existence of founder mutations simplifies the DNA-based screening programme, while the large number of individuals carrying these mutations strengthens the power of genotype/phenotype assessments. Data generated will be specific to the South African population and will ensure that the temptation to extrapolate from data collected from North American and European populations is avoided. This will facilitate improved patient management and counselling as it will allow for risk stratification and informed allocation of scarce resources in a local context.
In the present study we describe the mutation-screening programme followed to detect the molecular causes of HCM in South African patients and their immediate families. We also present the collation of HCM-causing mutations that have been found to date in this country and their relative contribution to the disease profile, together with a summary of their associated genotype/phenotype correlations. The relevance of these findings within the South African clinical setting is discussed in the context of new tenets and understanding relating to this insidious disease.

Materials and methods

Subjects and clinical examinations

This study was performed according to the guidelines of the Ethics Committee of the University of Stellenbosch. Informed consent was obtained from subjects, or from parents in the case of minors.

Index cases

A panel of 84 unrelated HCM-affected individuals, who had reported or had been referred to Tygerberg Hospital and who lived in the Western and Eastern Cape, was established. From available clinical records, these individuals were stratified into a subgroup of 37 patients with unequivocal HCM, and a second group of 47 patients in whom it was not possible to exclude other causes of hypertrophy. Consequently, the second category of individuals was referred to as the equivocal HCM-like subgroup.

Family studies

In cases where a DNA-based diagnosis was made, relatives of the index case (the proband) were traced and contacted whenever feasible. If they agreed to take part in the study, blood was drawn for DNA-based analysis (ascertainment of genotype); subsequently, only those family members who tested positive for a particular mutation were examined clinically in order to establish the clinical features associated with the mutation in that family (ascertainment of phenotype).

Clinical analysis

Clinical evaluations were as described previously. Briefly, 2-D and M-mode echocardiography were performed to estimate the extent and localisation of hypertrophy. Echocardiographic diagnosis of HCM was based on the presence of a maximal anterior interventricular septal thickness ≥ 13 mm. Standard 12-lead electrocardiography (ECG) was also performed, and electrocardiographic diagnosis was based on the presence of left ventricular hypertrophy (LVH) according to the point system of Romhilt and Estes or abnormal Q-waves. Other echocardiographic and ECG abnormalities compatible with HCM were also noted in the ascertainment of clinical status of mutation-carrying family members. Additionally, a history of disease-related and sudden cardiac death was established for each family.

Genetic analysis

Genomic DNA was extracted from peripheral blood lymphocytes as described previously. Thereafter, polymerase chain reaction (PCR)-based molecular genetic techniques, which allow for amplification of specific sarcomeric candidate genes and subsequent analysis of the selected DNA sequence for disease-causing mutations, were performed. The methods used in the study were PCR-single strand conformation polymorphism (PCR-SSCP) analysis, PCR-allele specific restriction enzyme analysis (ASREA) and direct sequencing, following standard protocols.

Mutation-screening programme

The mutation-screening programme that we follow is first to test for the three South African founder mutations (MYH7 Arg403Trp, Arg403Trp and Ala797Thr and TNNT2 Arg921Trp) in any proband newly entered into the study. If the results are negative, we concentrate on screening other regions of 7 of the different HCM-causing genes in which mutations have been previously described in South Africa, or elsewhere. This is because the presence of existing mutations indicates that these areas are of functional importance, where sequence variation may be deleterious. The complementary PCR-based methods used allow for detection of both known and novel mutations. Once a mutation is identified and characterised in a proband, a DNA-based diagnostic blood test is developed in order to genotype other known or HCM-affected panel members.

DNA-based diagnostic tests

Screening for the following mutations was performed by ASREA, using published protocols: MYH7 Arg249Gln, Arg403Trp, Ala797Thr, Arg719Gln and Glu499Lys, TNNT2 Arg921Trp and MYBPC3 Arg654His and Val896Met. The MYBPC3 Δc756 mutation was detected using PCR-SSCP analysis.

Genotype/phenotype analysis

Families with shared mutations were grouped together in order to make a clearer assessment of the clinical picture associated with each of the mutations. The maximum left ventricular wall thickness (LVWT) was established from the clinical evaluation data of individuals older than 16 years. Additionally, sudden and disease-related deaths in the affected families were used to construct Kaplan-Meier survival curves.

Results

Mutations detected

To date, DNA-based diagnosis has allowed for detection of 9 different disease-causing mutations among a total of 24 members of the panel of 84 South African HCM-affected probands (Table I). Of these, 21 of the mutation-bearing individuals were among the 37 probands assigned to the unequivocal HCM subgroup, while only 3 belonged to the subgroup of 47 individuals showing equivocal HCM-like disease.
Of the 5 different mutations in MYH7 detected in the panel, 3 (Arg249Gln, Arg403Trp and Arg719Trp) were among the more than 50 mutations in this gene that also occur elsewhere in the world,\(^2\) while 2 (Glu499Lys, Ala797Thr) were unique to South African HCM cases (Table I). None of the other numerous HCM-causing MYH7 mutations reported worldwide was present in members of the panel. Furthermore, the Ala797Thr and Arg403Trp mutations were detected in more than 1 member of the HCM-affected panel, and occurred in 10 and 2 apparently unrelated probands, respectively. It was possible to perform genotype/phenotype studies for 3 of the 5 MYH7 mutations, namely in 1 family bearing the Arg249Gln and another bearing the Arg403Trp mutation, and in 6 families carrying the Ala797Thr mutation. Among the families carrying the MYH7 Ala797Thr mutation, 1 was previously designated ped 101, in one branch of which, identified as 101a, were 2 of the original 84 probands, married to each other.\(^3\) These individuals both carried the MYH7 Ala797Thr mutation and genetic analysis indicated that they shared a common ancestor.\(^7\)

A TNNT2 mutation (Arg92Trp) previously reported in 2 probands\(^4\) was present in another 4 apparently unrelated probands, and clinical studies could be extended to 5 of these families. None of the 12 other reported TNNT2 mutations was present in the South African HCM panel.\(^2,3\)

In MYBPC3, a total of 3 different mutations (Arg654His, ΔC756, Val896Met), all unique to the South African population, were detected in 3 probands. Clinical studies could be extended to 2 families, 1 carrying the Arg654His and the other the Val896Met mutation. None of the 26 previously described mutations in MYBPC3 was present in other members of the panel.\(^2,3\)

In summary, therefore, 5 of the 9 HCM-causing mutations identified so far in MYH7 (2) and MYBPC3 (3) are unique to South African subpopulation groups. Three of the previously reported mutation occur in MYH7, while the TNNT2 Arg92Trp mutation was recently reported in a Japanese patient.\(^3\) Of the 3 mutations found in more than 1 proband, only the MYH7 Ala797Thr mutation occurred both in individuals of direct Caucasian descent and mixed ancestry, while the other 2 were found only in people of mixed ancestry. The remaining 6 mutations were represented equally among probands belonging to the 2 subpopulation groups (Table I). No new or previously described mutations were found in the areas of MYL2, MYL3, TNN1 or TPM genes investigated. Exons of ACTC and TNNT were not screened for mutations in the present study.

### Genotype/phenotype correlations

The localisation of hypertrophy associated with the different mutations varied, but the interventricular septum was inevitably affected whenever hypertrophy was present (Table II). The distribution of maximum LVWT in family members

---

**TABLE I. HCM-CAUSING MUTATIONS IDENTIFIED IN SOUTH AFRICAN PATIENTS**

<table>
<thead>
<tr>
<th>Causative gene</th>
<th>Mutation</th>
<th>Probands Unique to SA</th>
<th>Proband in HCM-group (N)</th>
<th>Proband in HCM-like group (N)</th>
<th>Ethnicity</th>
<th>Prognosis in SA population</th>
</tr>
</thead>
<tbody>
<tr>
<td>MYH7</td>
<td>Arg249Gln</td>
<td>No</td>
<td>1</td>
<td></td>
<td>MA</td>
<td>Good(^6)</td>
</tr>
<tr>
<td></td>
<td>Arg403Trp</td>
<td>No</td>
<td>2</td>
<td></td>
<td>MA</td>
<td>Good(^6)</td>
</tr>
<tr>
<td></td>
<td>Glu499Lys</td>
<td>Yes</td>
<td>1</td>
<td></td>
<td>Caucasian</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td>Ala797Thr</td>
<td>Yes</td>
<td>9</td>
<td></td>
<td>MA</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td>Arg719Trp</td>
<td>No</td>
<td>1</td>
<td></td>
<td>MA and</td>
<td>Good(^7)</td>
</tr>
<tr>
<td></td>
<td>Ala797Thr</td>
<td>Yes</td>
<td>1</td>
<td></td>
<td>Caucasian</td>
<td>Good(^8)</td>
</tr>
<tr>
<td>TNNT2</td>
<td>Arg92Trp</td>
<td>No</td>
<td>5</td>
<td>1</td>
<td>MA</td>
<td>Poor in males(^11)</td>
</tr>
<tr>
<td>MYBPC3</td>
<td>Arg654His</td>
<td>Yes</td>
<td>1</td>
<td></td>
<td>MA</td>
<td>Good(^5)</td>
</tr>
<tr>
<td></td>
<td>ΔC766</td>
<td>Yes</td>
<td>–</td>
<td>1</td>
<td>Caucasian</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td>Val896Met</td>
<td>Yes</td>
<td>1</td>
<td></td>
<td>Caucasian</td>
<td>Good(^4)</td>
</tr>
</tbody>
</table>

*With the exception of pedigree 101a.

SA = South African; MYH7 = beta myosin heavy chain gene; TNNT2 = cardiac troponin T gene; MYBPC3 = cardiac myosin binding protein C gene; MA = mixed ancestry; ND = not determined; PS = present study.

**TABLE II. LOCALISATION OF HYPERTROPHY IN MUTATION CARRIERS (PERCENTAGES OF CARRIERS OF DIFFERENT MUTATIONS SHOWING ECHOCARDIOGRAPHICALLY DETECTABLE HYPERTROPHY IN SPECIFIC CARDIAC REGIONS)**

<table>
<thead>
<tr>
<th>Area of hypertrophy</th>
<th>MYH7 (N = 7)</th>
<th>TNNT2 (N = 23)</th>
<th>MYBPC3 (N = 4)</th>
</tr>
</thead>
<tbody>
<tr>
<td>IVS only</td>
<td>57</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>IVS + PW</td>
<td>14</td>
<td>25</td>
<td>0</td>
</tr>
<tr>
<td>IVS + apex</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>IVS + PW + apex</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>No hypertrophy</td>
<td>29</td>
<td>64</td>
<td>75</td>
</tr>
</tbody>
</table>

MYH7 = beta myosin heavy chain gene; TNNT2 = cardiac troponin T gene; MYBPC3 = cardiac myosin binding protein C gene; IVS = anterior interventricular septum; PW = left ventricular posterior wall.

N = number of mutation carriers investigated.
carrying the different mutations clearly varied, with each mutation having a characteristic range (Fig. 1), mean and median values. Both the mean and the median maximum LVWT measurements were highest at 17 mm (standard deviation (SD) 8.6) and 13 mm, respectively, in the pooled families with the MYH7 Ala797Thr mutation (Table III). The lowest values were present in the families with the MYH7 Arg403Trp and TNNT2 Arg92Trp mutations, where the means of 10.5 mm (SD 4.1) and 11.0 mm (SD 5.4) and medians of 9.2 mm and 8.5 mm, respectively, fell below the 13 mm clinical cut-off line (Table III). In fact, the only other mutation associated with a mean value above 13 mm was MYH7 Arg249Gln at 15.5 mm (SD 6.4), although here too the median value was only 12 mm (Table III).

The percentages of mutation-carriers that could be diagnosed as HCM-affected by either the strict echocardiographic-diagnostic criterion alone, the strict ECG criteria alone, or both these criteria varied among the different mutations (Table IV). However, these diagnostic tools were insufficient to identify all individuals at risk of the consequences of their mutation status, and the number of carriers escaping clinical diagnosis by either echocardiography or ECG ranged between 43% and 75% (Table IV).

Survival curves indicated that the TNNT2 Arg92Trp mutation, despite being associated with mild to undetectable hypertrophy (Fig. 1 and Table II), had the poorest survival rate, with a precipitous drop in life expectancy between the ages of 15 and 26 years, and 50% survival by age 55 years (Fig. 2). On the other hand, most of the other mutations were associated with normal survival (Fig. 2). This was also generally true for families carrying the MYH7 Ala797Thr mutation, except for 1 family (ped 101a), in which the probands shared a common ancestor. In this case there was a high frequency of sudden death (Fig. 2).

![Fig. 1. Distribution of the maximum left ventricular wall thickness in carriers of the different mutations. The arrow indicates the 13 mm clinical diagnostic cut-off line. A797T rest = data from all families with the MYH7 Ala797Thr mutation; A797T ped = data from ped 101 bearing the MYH7 Ala797Thr mutation; A797T rest = data from all families with the MYH7 Ala797Thr mutation, excluding ped 101; R249Q = MYH7 Arg249Gln; R403W = MYH7 Arg403Trp; R292W = TNNT2 Arg92Trp; R654H = MYBPC3 Arg654His; V896M = MYBPC3 Val896Met; MLVWT (mm) = maximum left ventricular wall thickness in millimetres.](image)

**TABLE III. MEANS AND MEDIANS OF MAXIMUM LEFT VENTRICULAR WALL THICKNESS FOR THE DIFFERENT MUTATIONS**

<table>
<thead>
<tr>
<th>Gene and mutation</th>
<th>MYH7</th>
<th>TNNT2</th>
<th>MYPBC3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arg249Gln (N = 7)</td>
<td>15.5 ± 6.4</td>
<td>10.5 ± 4.1</td>
<td>17 ± 8.6</td>
</tr>
<tr>
<td>Arg403Trp (N = 20)</td>
<td>10.5 ± 4.1</td>
<td>17 ± 8.6</td>
<td>11 ± 5.4</td>
</tr>
<tr>
<td>Ala797Thr (N = 24)</td>
<td>17 ± 8.6</td>
<td>11 ± 5.4</td>
<td>11.7 ± 6.6</td>
</tr>
<tr>
<td>Arg92Trp (N = 23)</td>
<td>11 ± 5.4</td>
<td>11.7 ± 6.6</td>
<td>10.8 ± 4.1</td>
</tr>
<tr>
<td>Arg654His (N = 4)</td>
<td>11.7 ± 6.6</td>
<td>10.8 ± 4.1</td>
<td></td>
</tr>
<tr>
<td>Val896Met (N = 4)</td>
<td>10.8 ± 4.1</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*MYH7 = beta myosin heavy chain gene; TNNT2 = cardiac troponin T gene; MYPBC3 = cardiac myosin binding protein C gene; N = number of mutation carriers investigated; SD = standard deviation.

**TABLE IV. PERCENTAGE OF MUTATION-CARRIERS MEETING CLINICAL DIAGNOSTIC CRITERIA FOR HCM**

<table>
<thead>
<tr>
<th>Gene and mutation</th>
<th>MYH7</th>
<th>TNNT2</th>
<th>MYPBC3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arg249Gln (N = 7)</td>
<td>43</td>
<td>43</td>
<td>43</td>
</tr>
<tr>
<td>Arg403Trp (N = 20)</td>
<td>43</td>
<td>43</td>
<td>43</td>
</tr>
<tr>
<td>Ala797Thr (N = 24)</td>
<td>43</td>
<td>43</td>
<td>43</td>
</tr>
<tr>
<td>Arg92Trp (N = 23)</td>
<td>43</td>
<td>43</td>
<td>43</td>
</tr>
<tr>
<td>Arg654His (N = 4)</td>
<td>25</td>
<td>25</td>
<td>25</td>
</tr>
<tr>
<td>Val896Met (N = 4)</td>
<td>25</td>
<td>25</td>
<td>25</td>
</tr>
</tbody>
</table>

*Positive diagnosis: Echocardiography*: 43/43/43/43 for MYH7/TNNT2/MYPBC3/both; ECG*: 43/43/43/43 for MYH7/TNNT2/MYPBC3/both. Negative diagnosis: Echocardiography and ECG:

*MYH7 = beta myosin heavy chain gene; TNNT2 = cardiac troponin T gene; MYPBC3 = cardiac myosin binding protein C gene; N = number of mutation carriers investigated.*
Importance of rigorous initial clinical diagnosis

Molecular-based diagnosis identified sarcomeric protein-encoding genes as the cause of hypertrophy in 57% (21/37) of patients assigned to the subgroup of 37 individuals with unequivocal HCM. In the second subgroup, with HCM-like disease, a molecular cause was found only in 6% (3/47) of patients. The less than 100% success rate in the former unequivocal group reflects the high genetic heterogeneity of the disease, and it may be that these individuals carry mutations in regions of the candidate genes other than those that were investigated in the present study. In addition, exons of ACTC and ITN in which HCM-causing mutations have been described were not screened,\(^{2,9}\) while other HCM-causative genes may yet be unidentified.

The higher success rate in the subgroup with the clinically certain diagnosis compared with that of the equivocal HCM-like subgroup raises questions about the causes of hypertrophy in the latter group. It emphasises the importance of a rigorous clinical diagnosis in probands, viz., strictly adhering to the definition of HCM as idiopathic hypertrophy that occurs in the absence of hypertension, syndromic conditions, diabetes or valvular heart disease, before proceeding with DNA-based diagnosis of an index case. Without clear clinical diagnosis in the probands, screening for the causal mutation is likely to be prolonged and the success rate may be low. This has cost implications, both to the service and research-based components of our molecular diagnosis programme. Furthermore, from a clinical perspective the successful identification of a disease-causing mutation in a proband means that molecular diagnosis can also be offered to first-degree family members, thereby identifying truly at-risk individuals, and allowing for the implementation of appropriate management strategies for all mutation carriers. If the mutation is new to South Africa, research-based genotype/phenotype correlations can be instigated to serve as a further aid to patient care and counselling.

South African HCM-causing mutations

Among the 9 different disease-causing mutations detected in the HCM panel to date, only 4 that have been described elsewhere were present. This supports previously held views that the specific molecular causes of HCM will be unique to any geographical area.\(^{23}\) In conjunction with caveats concerning extrapolation of data obtained from other populations to those in South Africa,\(^{4}\) our results underscore the need to identify the spectrum of genetic causes in our country and establish their clinical impact in a local context.

Thereafter, mutation-screening can be used with confidence to facilitate diagnosis and prognostication of patients and their families in South Africa.

We have previously shown that the MYH7 Ala797Thr and Arg403Trp and the TNNT2 Arg92Trp mutations, which account for a substantial number of HCM cases in apparently unrelated probands, at least in the Western and Eastern Cape, are the result of founder effects.\(^{4}\) The MYH7 Ala797Thr founder mutation is the cause of 25% of the cases of HCM among members of the unequivocally diagnosed subgroup, the MYH7 Arg403Trp mutation for 5% of cases and the TNNT2 Arg92Trp mutation for 11% of cases. These findings are beneficial, both to the molecular and clinical programmes, as it is logical first to test for these mutations before embarking on a detailed search for other possible molecular causes, which can be time-consuming. Furthermore, owing to the large patient base in which these mutations could be studied, their genotype/phenotype correlations provided a reliable foundation for prognostication.

Hypertrophy and sudden death

In the overwhelming majority of cases in which it was feasible to investigate family members, at least 1 mutation carrier other than the proband was identified, indicating the strong familiar nature of HCM.\(^{4,18,36,37}\) Therefore, investigations should not end with diagnosis of HCM in the proband, not even if that diagnosis was made post mortem. In fact, in such cases it is probably especially relevant to continue with a family follow-up, as some mutations are associated with higher risk of sudden cardiac death than others (Fig. 2). It should also be borne in mind that numerous studies,\(^{12,17,36,37,39}\) including our own, have identified many mutation-carrying relatives of a proband who did not demonstrate any clinical signs of the disease, even when well past puberty. In every family studied there was a striking number...
of mutation carriers that did not meet the clinically defined criteria for HCM (Table III). The most clinically significant information generated is probably the finding that there was no evidence of cardiac hypertrophy in half of the individuals bearing the TNNT2 Arg92Ttp mutation, which is nevertheless associated with reduced survival (Fig. 2). Although a higher percentage of MYH7 Arg403Ttp carriers did not exhibit hypertrophy (60%), this mutation is considered benign. The later age of onset of HCM associated with MYBPC3 mutations probably accounts for the highest values of 75% of MYBPC3 Arg654His and VAl896Met carriers not exhibiting hypertrophy. However, all these individuals, in whom the disease can only be identified by molecular means, are still at risk of the clinical consequences of their mutations, which may be very dire.

The fact that hypertrophy and the risk of sudden death are independent features of HCM is illustrated especially well by the founder mutations. Genotype/phenotype correlations revealed that both the MYH7 Ala797Thr and Arg403Ttp founder mutations are generally associated with good survival, despite the tendency towards more overt hypertrophy in the case of the MYH7 Ala797Ttp mutation. An exception to the favourable prognosis associated with the latter mutation was noted in the offspring of the marriage of two mutation carriers who shared common ancestry in a branch of family 101. It can be speculated that this common genetic background might have introduced two copies of other deleterious modulating genes, which together may have exacerbated susceptibility to the main MYH7 Ala797Thr disease-causing mutation in the younger generation. On the other hand, the TNNT2 Arg92Ttp founder mutation is invariably associated with poor survival, especially among males, despite the fact that echocardiographically detectable hypertrophy is very often absent in these patients.

It is therefore clear that molecular diagnosis offers several benefits as an adjunct to clinical diagnosis. Firstly, once the mutation has been identified in the clinically diagnosed proband, molecular diagnosis facilitates unequivocal identification, regardless of age or current clinical diagnostic status, of those family members truly at risk from the disease. This leads to savings and focused management, as only those patients carrying the mutation require clinical follow-up. Secondly, well-established genotype/phenotype correlations, which are made possible by a combination of clinical assessment and molecular diagnosis, allow informed prognostication, thereby aiding realistic counselling for a particular affected family and for other patients carrying the same mutation. Thirdly, knowledge of the specific causal mutation and its associated phenotype allows for risk stratification of patients, so that only those individuals carrying a mutation that specifically confers a high risk of sudden death, such as the TNNT2 Arg92Ttp mutation, need to be considered for costly interventions. It has been recommended that patients with mutations in TNNT2 receive automatic implantable cardioverter defibrillators; however, such intervention is generally too costly for the South African health care system. Consequently, patients, especially males, with the TNNT2 Arg92Ttp mutation are treated from 12 years of age with low doses of amiodarone, which has been reported to reduce the likelihood of sudden death in HCM. These patients are also actively discouraged from participating in any type of sport. Patients with low-risk mutations are treated symptomatically; it is recommended that they do not participate in intensive sport.

Finally, an important aim of our investigations is to identify the full spectrum of HCM-causing mutations and their clinical course in all population groups in South Africa, and we will continue to search prospectively for new index cases. It is becoming clear that while the presence of HCM-causing mutations is higher than previously suspected, the clinical consequences of bearing these defects require careful interpretation. Worldwide, efforts to characterise disease-causing mutations are contributing to an understanding of the underlying molecular pathophysiology of familial HCM, which will in turn lead to the development of new, rational management and treatment strategies for this clinically and genetickly heterogeneous condition.

Since going to press, the recent sudden cardiac death of an individual carrying the MYH7 Ala797Thr mutation has been recorded. This patient had severe hypertrophy (LVWT > 35 mm).

References


30. Rothhilt DW, Estes EH. A point score system for the ECG diagnosis of left ventricular hypertrophy. Am Heart J 1968; 75: 752-758.


