Studies in Cardiomyopathy:

Looking beyond the familiar

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Dedication

I dedicate this work to my parents, Tembeka and Bubele Jamba, without whose support, love and encouragement it would not have been possible.

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Declaration

The work in this thesis is entirely my own.

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Abbreviations

ACE = Angiotensin converting enzyme

AD = Autosomal dominant

AIDS = Acquired immunodeficiency syndrome

AHA = American Heart Association

AR = Autosomal recessive

ARB = Angiotensin receptor blocker

ARVC = Arrhythmogenic right ventricular cardiomyopathy

BMI = Body mass index

BSA = Body surface area

CAD = Coronary artery disease

CMR = Cardiovascular magnetic resonance

CRT = Cardiac resynchronization therapy

CVD = Cardiovascular disease

DCM = Dilated cardiomyopathy

ECG = Electrocardiography

EMB = Endomyocardial biopsy

EMF = Endomyocardial fibrosis

ESC = European Society of Cardiology

FDC = Familial dilated cardiomyopathy

HCM = Hypertrophic cardiomyopathy

HHFP = Hypertensive heart failure of pregnancy

HIV = Human immunodeficiency virus

ISFC = International Society and Federation of Cardiology

IVS = Interventricular septum

IVSd = Interventricular septum in diastole

LA = Left atrium

LBBB = Left bundle branch block

LGE = Late gadolinium enhancement

LV = Left ventricle/ventricular

LVEDD = Left ventricular end-diastolic dimension

LVEDV = Left ventricular end-diastolic volume

LVEF = Left ventricular ejection fraction

LVESV = Left ventricular end-systolic volume

LVH = Left ventricular hypertrophy

LVMI = Left ventricular mass indexed to body surface area

LVOT = Left ventricular outflow tract

LVPFW = Left ventricular posterior free wall

LVSV = Left ventricular stroke volume

LVNC = Left ventricular non-compaction

MC = Mutation carrier

NYHA = New York Heart Association

OHT = Orthotopic heart transplantation

PIH = Pregnancy induced hypertension

PPCM = Peripartum cardiomyopathy

SCD = Sudden cardiac death

SLE = Systemic lupus erythematosus

RBBB = Right bundle branch block

SSA = Sub-Saharan Africa

SSc = Systemic sclerosis

RA = Rheumatoid arthritis

RCM = Restrictive cardiomyopathy

RV = Right ventricle/ventricular

WHO = World Health Organization

X-LR = X-linked recessive

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Published articles

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Abstract

Background: Little is known about the mechanisms, clinical characteristics, natural history and outcomes of cardiomyopathy amongst Africans. Familial aggregation of cardiomyopathy has not been studied systematically in an African setting. Further, it is not clear whether the various phenotypic expressions of cardiomyopathy represent disparate clinical entities, or whether they are merely different forms of the same disease manifested differently in different circumstances.

Methods: Two cohorts of patients with cardiomyopathy were utilised for this study: (1) patients with cardiomyopathy seen at the specialist cardiomyopathy clinic at Groote Schuur Hospital, Cape Town between February 1, 1996 and December 31, 2009; and (2) a group of hypertrophic cardiomyopathy (HCM) patients and first degree relatives seen in a specialist cardiogenetic clinic at Tygerberg Hospital, who underwent cardiovascular magnetic resonance (CMR) imaging at Groote Schuur Hospital.

Results: 29 out of 109 unrelated cases with dilated cardiomyopathy (DCM) had familial disease, and had a significantly younger mean age of onset of cardiomyopathy than non-familial cases. Two of the 29 patients with familial DCM had at least one relative who was diagnosed with peripartum cardiomyopathy (PPCM). Pedigree analysis of the 29 families was consistent with autosomal dominant (AD) inheritance in 72.4%, autosomal recessive inheritance in 17.2% and X-linked recessive inheritance in 10.4%. Cardiac chambers were significantly more dilated with poorer left ventricular (LV) systolic function in idiopathic than familial cases. The mortality rate of 40% after a median follow-up of 5 years was, however, similar in both groups. The presence of New York Heart Association functional class III and IV symptoms was an independent predictor of mortality, whilst heart transplantation was an independent predictor of survival in both groups. Digoxin use

without serum monitoring was a significant predictor of mortality in idiopathic DCM. We present the longest follow-up study of PPCM in Africans, showing (1) a case fatality rate of 16.7% over a median of 4.3 years in patients on modern medical therapy; (2) PPCM is associated with adverse outcomes; (3) chronic heart failure occurs in 80% of patients studied, and with virtually no normalisation of LV dysfunction; and (4) that persistence of symptoms of heart failure are the strongest predictor of death in patients with PPCM. We compared the time of onset of symptoms, clinical profile and outcome of patients with hypertensive heart failure of pregnancy (HHFP) and PPCM and found onset of symptoms to be postpartum in all PPCM patients and antepartum in 85% of HHFP. PPCM was associated with more severe complications and a higher rate of mortality than HHFP.

The clinical presentation of HCM was similar to that reported in other studies. The South African founder mutations that cause HCM were not found in the 42 probands studied and 29% of those tested for mutations in 15 genes had disease causing mutations in *MYH7* and *MYBPC3*. Overall survival of HCM was 74% at 10 years, which was similar to the general South African population. The independent predictor of mortality was New York Heart Association (NYHA) functional class at last visit (P=0.026), but not by presence of a disease-causing mutation (P=0.474). Finally, we investigated the correlation between genotype and cardiovascular magnetic resonance (CMR) phenotype in HCM patients with the 3 founder South African mutations and found variable phenotypic expression associated with the different genotypes.

Conclusions: Familial DCM affects at least a quarter of African patients with DCM, presents at a young age, is associated with PPCM, and follows an AD pattern of inheritance in the majority of families. Patients with idiopathic DCM have greater cardiac dysfunction than those with familial disease, but mortality remains similarly high in both groups. Digoxin use without drug-level monitoring may be associated with increased mortality in idiopathic DCM. Persistent symptoms predict poor outcomes in PPCM. There are significant differences in the time of onset of heart

failure, clinical and echocardiographic features, and outcome of HHFP compared to PPCM, indicating that the presence of hypertension in pregnancy-associated heart failure may not fit the case definition of idiopathic PPCM. Comprehensive genetic screening was associated with a 29% yield of causal genetic mutations in South African HCM cases, all in *MYH7* and *MBPC3* genes. NYHA functional class at last visit, but not presence of a disease-causing mutation, was an independent predictor of mortality. Multiparametric CMR demonstrated variable phenotypic expression in HCM mutation carriers with differing genotypes.

Keywords

Cardiomyopathy, sub-Saharan Africa, South Africa, heart failure, dilated cardiomyopathy, familial cardiomyopathy, hypertrophic cardiomyopathy, peripartum cardiomyopathy, cardiovascular magnetic resonance, echocardiography

Chapter 1

Introduction and study rationale

1.1 Historical perspectives, evolving concepts and definition of cardiomyopathy

Cardiomyopathies are primary disorders of heart muscle associated with left ventricular (LV) dysfunction and development of heart failure.[1] The very first description of dilated cardiomyopathy was probably provided by Gillanders in 1951. In 1980, the World Health Organization (WHO) defined cardiomyopathies as "heart muscle diseases of unknown cause," to distinguish cardiomyopathy from cardiac dysfunction due to known entities such as hypertension, ischaemic heart disease, valvular disease, pericardial disease or congenital heart disease.[2] However, in clinical practice, the term "cardiomyopathy" has also often been applied incorrectly to diseases of known cause (e.g. '*ischaemic cardiomyopathy*' and '*hypertensive cardiomyopathy*'). Consequently, the 1995 WHO/International Society and Federation of Cardiology (ISFC) Task Force on the Definition and Classification of the Cardiomyopathies expanded the classification to include all diseases affecting heart muscle and to consider aetiology as well as the dominant pathophysiology.[3] In this 1995 classification, the cardiomyopathies were defined as "diseases of the myocardium associated with cardiac dysfunction." They were classified according to anatomy and pathophysiology into the following types, each of which has multiple different causes: (a) dilated cardiomyopathy (DCM); (b) hypertrophic cardiomyopathy (HCM); (c) restrictive cardiomyopathy (RCM); (4) arrhythmogenic right ventricular cardiomyopathy (ARVC); and (5) unclassified cardiomyopathies.[3]

In 2006, the American Heart Association (AHA) published a scientific statement that proposed a contemporary definition and classification of the cardiomyopathies.[4] The AHA expert consensus panel proposed the following definition: "Cardiomyopathies are a heterogeneous group of diseases of the myocardium associated with mechanical and/or electrical dysfunction that usually (but not invariably) exhibit inappropriate ventricular hypertrophy or dilatation and are due to a variety of causes that frequently are genetic. Cardiomyopathies either are confined to the heart or are part of a generalised systemic disorders, often leading to cardiovascular death or progressive heart failure-related disability."[4] In this new scheme, cardiomyopathies were categorised into two groups: (a) primary cardiomyopathies, which may be genetic, mixed (genetic or non-genetic), or acquired; and (b) secondary cardiomyopathies, which are accompanied by other organ/system involvement (Table 1.1). The AHA definition and classification were not intended to provide methodologies for clinical diagnosis, but were rather a scientific scheme that aimed to aid in the understanding of this complex group of disorders. The main departure of the proposed AHA Scientific Statement definition from previous classifications was the inclusion of the ion channelopathies as primary cardiomyopathies, despite the absence of gross structural abnormalities of cardiac muscle.

In 2007, the European Society of Cardiology's (ESC) working group on myocardial and pericardial diseases presented an update to the WHO/ISFC classification in which cardiomyopathy was defined as: "A myocardial disorder in which the heart muscle is structurally and functionally abnormal in the absence of coronary artery disease, hypertension, valvular disease and congenital heart disease sufficient to explain the observed myocardial abnormality." This ESC classification was a significant departure from the WHO/ISFC classification in that the distinction between familial/genetic and non-familial/non-genetic causes of LV dysfunction were emphasised, and heart disease secondary to hypertension, ischaemic heart disease, pericardial, valvular, and congenital

disorders were not included.[5] Importantly, in the ESC classification, unlike in the AHA classification, channelopathies were excluded as well (Figure 1.1 and Table 1.2).

Table 1.1. The AHA classification of the cardiomyopathies

Primary cardiomyopathies	Genetic	HCM, ARVC, LVNC, conduction defects, mitochondrial myopathies, ion channel disorders
	Mixed	DCM, RCM
	Acquired	Inflammatory, Takotsubo, peripartum, tachycardia-induced, infants of diabetic mothers
Secondary cardiomyopathies	Infiltrative	Amyloidosis, Gaucher's, Hurler's, Hunter's
	Storage	Anderson-Fabry's, glycogen storage diseases, Niemann-Pick disease, haemochromatosis
	Toxicity	Drugs, heavy metals
	Endomyocardial	Endomyocardial fibrosis, Loeffler's endocarditis
	Inflammatory	Sarcoidosis
	Endocrine	Diabetes, hyperthyroidism, hypothyroidism, hyperparathyroidism, phaeochromocytoma, acromegaly
	Cardiofacial	Noonan's, lentoginosis
	Neuromuscular	Friedrich's ataxia, Duchenne-Becker muscular dystrophy, myotonic dystrophy
	Nutritional	Beriberi, scurvy, selenium deficiency
	Autoimmune	RA, SLE, SSc, dermatomyositis
	Consequence of therapy	Anthracyclines, radiation, cyclophosphamide

ARVC, arrhythmogenic right ventricular cardiomyopathy; DCM, dilated cardiomyopathy; HCM, hypertrophic cardiomyopathy; LVNC, left ventricular non-compaction; RA, rheumatoid arthritis; RCM, restrictive cardiomyopathy; SLE, systemic lupus erythematosus; SSc, systemic sclerosis

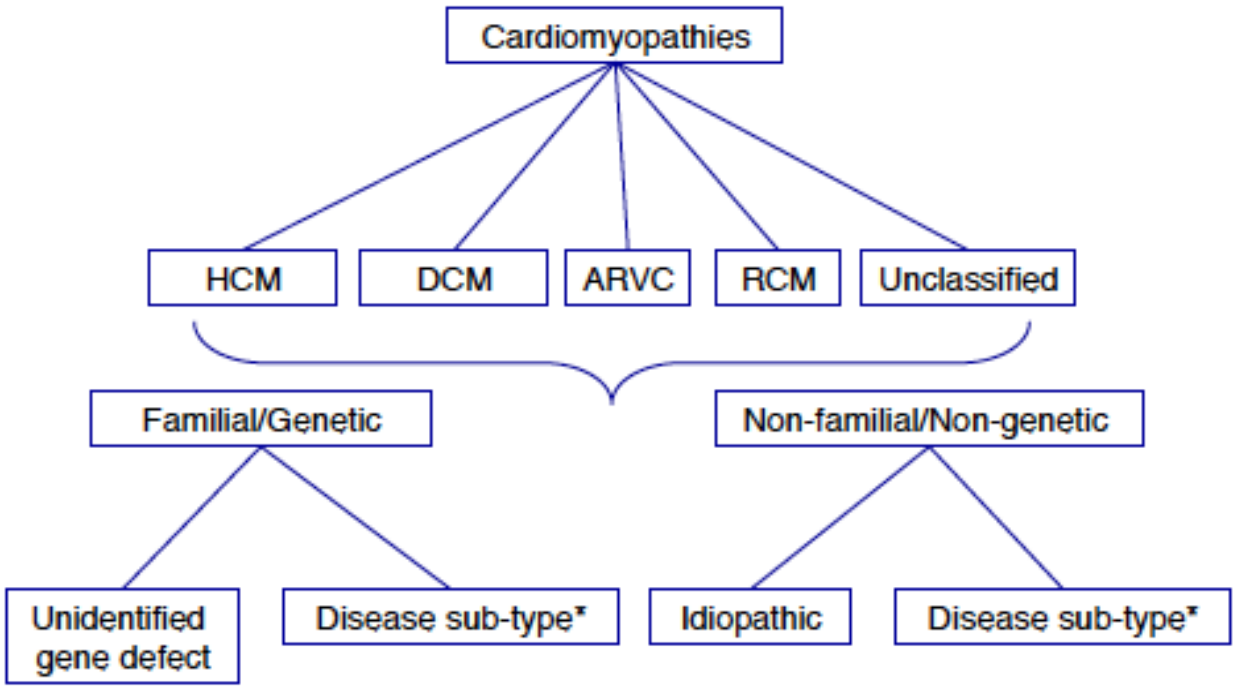


Figure 1.1. European Working Group on Myocardial and Pericardial Diseases ESC classification of the cardiomyopathies
ARVC, arrhythmogenic right ventricular cardiomyopathy; DCM, dilated cardiomyopathy; HCM, hypertrophic cardiomyopathy; RCM, restrictive cardiomyopathy
From Elliott P, et al. Classification of the cardiomyopathies: a position statement from the European Society of Cardiology Working Group on Myocardial and Pericardial diseases. *Eur Heart J* 2008;29:270-276.

In 2013, the World Heart Federation endorsed the MOGE(S) classification [6,7] which was developed from the need to describe cardiomyopathies by integration of a morphofunctional phenotype-based description with information regarding extracardiac organ involvement and clinical (pattern of inheritance) and molecular (disease gene and mutation) genetics in familial disease. The MOGE(S) classification also aimed to describe sporadic cardiomyopathies, and specify their etiology when known or unknown (Table 1.3).

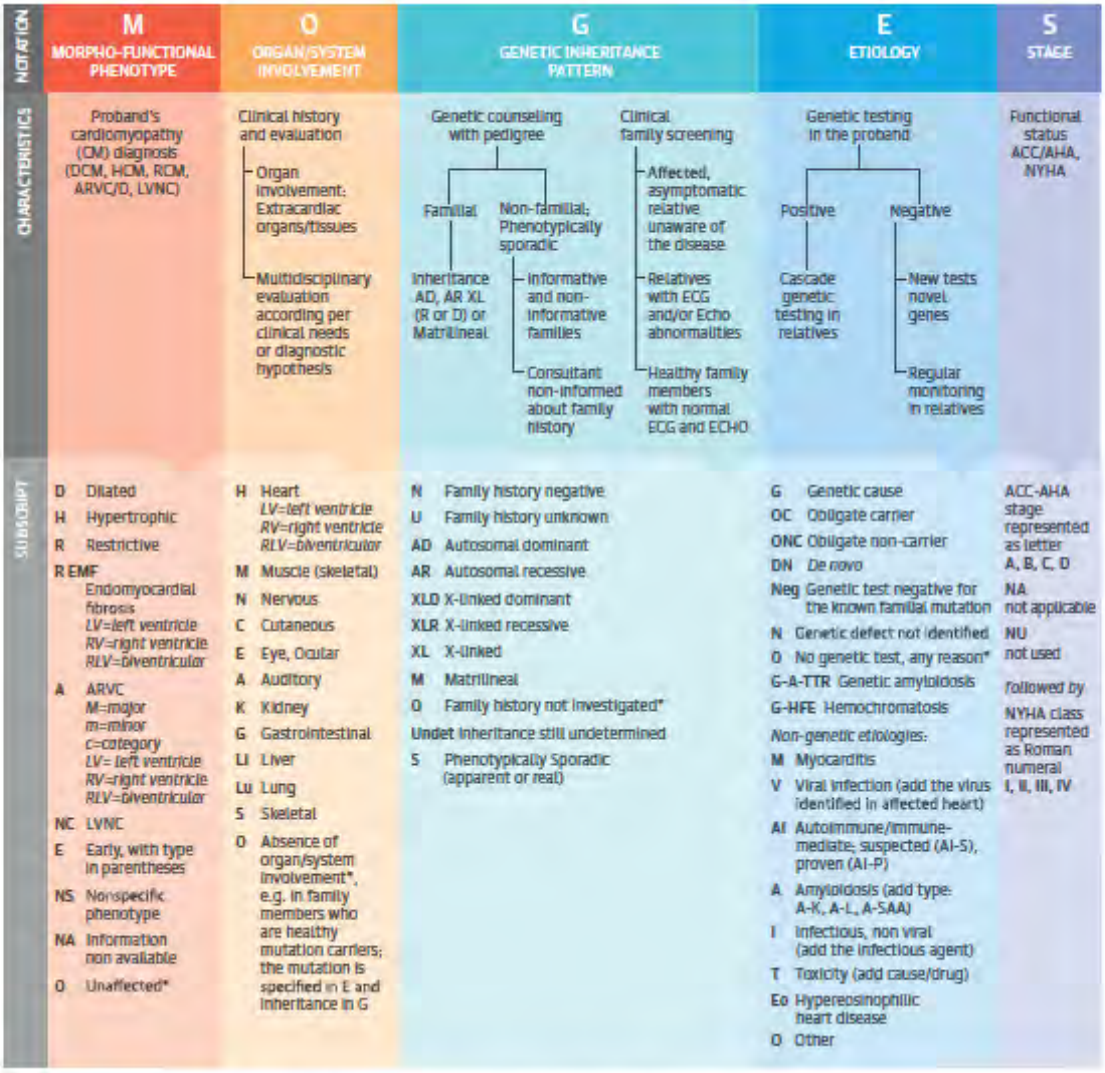
Table 1.2. The ESC classification of cardiomyopathies by cause

	HCM	DCM	ARVC	RCM	Unclassified
Familial	Familial, unknown gene	Familial, unknown gene	Familial, unknown gene	Familial, unknown gene	Left ventricular non-compaction
	Sarcomeric protein mutations	Sarcomeric protein mutations (see HCM)	Intercalated disc protein mutations	Sarcomeric protein mutations	
	β myosin heavy chain			Troponin I (RCM ++/− HCM)	Barth syndrome
	Cardiac myosin binding protein C	Z-band	Plakoglobin	Essential light chain of myosin	Lamin A/C
	Cardiac troponin I	Muscle LIM protein	Desmoplakin	Familial amyloidosis	ZASP
	Troponin-T	TCAP	Plakophilin 2	Transthyretin (RCM + neuropathy)	α-dystrobrevin
	α-tropomyosin	Cytoskeletal genes	Desmoglein 2	Apolipoprotein (RCM + nephropathy)	
	Essential myosin light chain	Dystrophin	Desmocollin 2	Desminopathy	
	Regulatory myosin light chain	Desmin	Cardiac ryanodine receptor (RyR2)	Pseuxanthoma elasticum	
	Cardiac actin	Metavinculin		Haemochromatosis	
	α-myosin heavy chain	Sarcoglycan complex	Transforming growth factor-β3 (TGFβ3)	Anderson–Fabry disease	
	Titin	CRYAB		Glycogen storage disease	
	Troponin C	Epicardin			
	Muscle LIM protein	Nuclear membrane			
	Glycogen storage disease (e.g. Pompe PRKAG2, Forbes', Danon)	Lamin A/C			
		Emerin			
	Lysosomal storage diseases (e.g. Anderson–Fabry, Hurler's)	Mildly dilated CM			
		Intercalated disc protein mutations (see ARVC)			
	Disorders of fatty acid metabolism				
	Carnitine deficiency	Mitochondrial cytopathy			
	Phosphorylase B kinase deficiency				
	Mitochondrial cytopathies				
	Syndromic HCM				
	Noonan's syndrome				
	LEOPARD syndrome				
	Friedreich's ataxia				
	Beckwith–Wiedemann syndrome				
	Swyer's syndrome				
	Other				
	Phospholamban promoter				
	Familial amyloid				
Non-familial	Obesity	Myocarditis (infective/toxic/immune)	Inflammation?	Amyloid (AL/prealbumin)	Tako Tsubo cardiomyopathy
	Infants of diabetic mothers	Kawasaki disease		Scleroderma	
	Athletic training	Eosinophilic (Churg Strauss syndrome)		Endomyocardial fibrosis	
	Amyloid (AL/prealbumin)			Hypereosinophilic syndrome	
		Viral persistence		Idiopathic	
		Drugs		Chromosomal cause	
		Pregnancy		Drugs (serotonin, methysergide, ergotamine, mercurial agents, busulfan)	
		Endocrine			
		Nutritional — thiamine, carnitine, selenium, hypophosphataemia, hypocalcaemia		Carcinoid heart disease	
				Metastatic cancers	
		Alcohol		Radiation	
		Tachycardiomyopathy		Drugs (anthracyclines)	

ARVC, arrhythmogenic right ventricular cardiomyopathy; DCM, dilated cardiomyopathy; HCM, hypertrophic cardiomyopathy; RCM, restrictive cardiomyopathy

From Elliott P, et al. Classification of the cardiomyopathies: a position statement from the European Society of Cardiology Working Group on Myocardial and Pericardial diseases. *Eur Heart J* 2008;29:270-276.

Table 1.3. The MOGE(S) classification of cardiomyopathies



CENTRAL ILLUSTRATION **The MOGE(S) Nosology System for Classifying CM Patients**

Evaluation of cardiomyopathy patients and development of MOGE(S) nosology. **(M)** The morphofunctional phenotype description may contain more information using standard abbreviations: AVB = atrioventricular block; LQT = prolongation of the QT interval; ↓PR = short PR interval; ↓R = low electrocardiographic voltages; WPW = Wolf Parkinson White syndrome; and other clinical red flags. These red flags are to be placed in parentheses after the notation of morphofunctional phenotype. *Overlapping (H+R), (D+A), (NC+H), (H+D), (D+NC) or more complex combinations such as (H+R+NC).* *Notation is zero (0) not the letter "O." **(E)** The etiologic annotation provides the description of the specific disease gene and mutation, as well as a description of nongenetic etiology. Even when genetic analysis is not available, the **(G)** may inform about a genetic disease, supporting family monitoring strategies. #According to the Human Genome Variation Society, genetic variants should be classified based on their effects on gene function as: affecting function, probably affecting function, unknown (variants of unknown significance [VUS]), probably not affecting function, and not affecting function. A color code assigned to each variant can provide information about the potential role of the identified variant: affects function or probably affects function **(red)**; Variant of Unknown Significance (VUS) **(yellow)**; and probably does not affect function (or probably no functional effect) or does not affect function (no functional effect) **(green)**. The compilation is guided by the MOGES app (63). ACC = American College of Cardiology; AHA = American Heart Association; ARVC/D = arrhythmogenic right ventricular cardiomyopathy/dysplasia; DCM = dilated cardiomyopathy; ECG = electrocardiogram; ECHO = echocardiogram; HCM = hypertrophic cardiomyopathy; LVNC = left ventricular noncompaction; NYHA = New York Heart Association; RCM = restrictive cardiomyopathy.

From Arbustini E, et al. The MOGE(S) classification for a phenotype-genotype nomenclature of cardiomyopathy: endorsed by the World Heart Federation. *J Am Coll Cardiol* 2013;62:2046-2072.

In 2014, the ESC published guidelines on the diagnosis and management of hypertrophic cardiomyopathy.[8] These guidelines comprise a comprehensive review of the literature on HCM, the evolving understanding of the disease, current concepts, diagnosis of HCM and HCM phenocopies and evidence based strategies for risk stratification and management of the condition. HCM is defined by the presence of increased left ventricular (LV) wall thickness that is not solely explained by abnormal loading conditions (Figure 1.2).

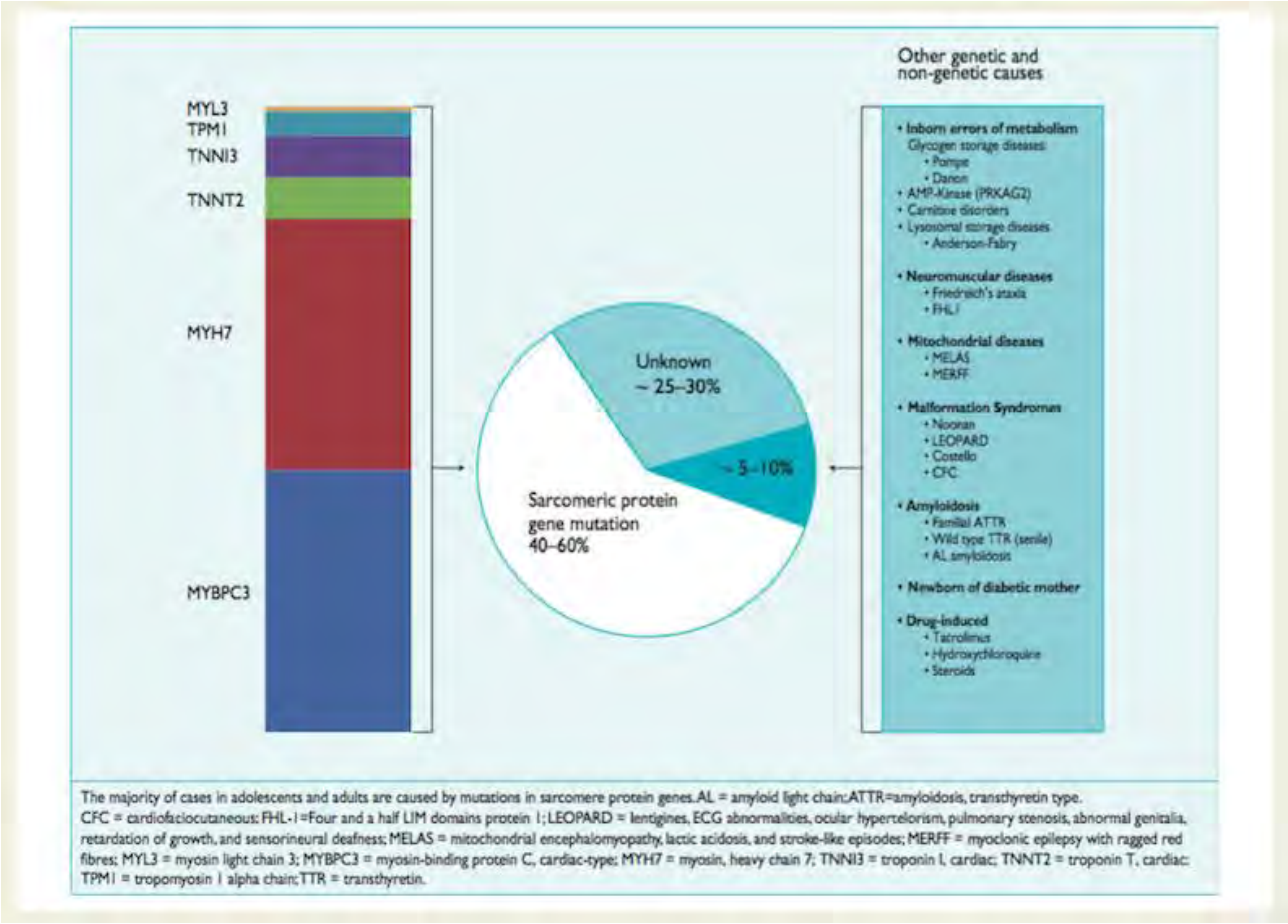


Figure 1.2. The complex aetiology of left ventricular hypertrophy
From Elliot P, et al. 2014 ESC Guidelines on diagnosis and management of hypertrophic cardiomyopathy: the Task Force for the Diagnosis and Management of Hypertrophic Cardiomyopathy of the European Society of Cardiology (ESC). *Eur Heart J* 2014;35:2733-2779.

In 1994, an International Task Force proposed criteria for the clinical diagnosis of ARVC that facilitated recognition and interpretation of the frequently nonspecific clinical features of ARVC, resulting in confirmatory clinical diagnosis in index cases through exclusion of phenocopies and

provision of a standard on which clinical research and genetic studies could be based.[9] Structural, histological, electrocardiographic, arrhythmic, and familial features of the disease were incorporated into the criteria, subdivided into major and minor categories according to the specificity of their association with ARVC. At that time, clinical experience with ARVC was dominated by symptomatic index cases and sudden cardiac death (SCD), which represented the severe end of the disease spectrum and only the 'tip of the iceberg'. Consequently, the 1994 criteria were highly specific but lacked sensitivity for early and familial disease.

In 2010, a new set of Task Force criteria were proposed,[10,11] representing a working framework to improve the diagnosis and management of ARVC which relies on the demonstration of structural, functional, and electrophysiological abnormalities that are caused by or reflect the underlying histological changes. Technical advances in cardiovascular magnetic resonance (CMR) and 2-dimensional echocardiography have improved the capability to image the right ventricle (RV) with reproducible measurements of volume and systolic function, which permits classification of severity and differentiation from normality. Previous diagnostic reliance on subjective assessment of RV wall thinning, wall motion abnormalities, and fatty infiltration of the myocardium by CMR has proven problematic, and has consequently been modified in the revise criteria. Recognition of significant fatty involvement without concomitant fibrosis of the RV in normal individuals renders this unique CMR capability of limited value. Late gadolinium enhancement (LGE) on CMR permits myocardial tissue characterisation in both the LV and RV. It can be difficult to be certain of late enhancement for characterisation of RV myocardium because of the thin wall of the RV and possible confusion with fat.

1.2 Epidemiology of cardiomyopathy in Africa

Heart failure has emerged as a dominant form of cardiovascular disease (CVD) in Africa, and holds great social and economic relevance because of its high prevalence, high mortality, and impact on young economically active individuals.[12] The causes of heart failure in Africans remain, largely, non-ischaemic. Hypertension, cardiomyopathy, rheumatic heart disease, chronic lung disease and pericardial disease are the main contributors to the aetiology of cardiac failure in sub-Saharan Africa (SSA), accounting for over 90% of cases.[12] In SSA, endemic cardiomyopathies include DCM, peripartum cardiomyopathy (PPCM), and endomyocardial fibrosis (EMF). Non-endemic cardiomyopathies apparently occur with the same frequency as in other parts of the world, and include HCM, ARVC and human immunodeficiency virus (HIV)-associated cardiomyopathy. Unlike in the developed world where the epidemiology of cardiomyopathy has been studied and described, there are no population-based incidence and prevalence studies of cardiomyopathy from Africa and other developing regions of the world. Hence, information on the epidemiology of cardiomyopathy in SSA is obtained, largely, from hospital-based studies.[12]

Idiopathic dilated cardiomyopathy (DCM): In South Africa and Uganda, DCM accounts for 10-17% of all cardiac conditions encountered at autopsy.[13-15] In SSA, DCM occurs commonly in the third and fourth decades of life, with men affected twice as commonly as women. Two thirds of African patients with DCM, especially those who are more than 55 years of age, have persistently low arterial blood pressure, ventricular arrhythmias, and/or atrioventricular valve incompetence and die within 5 years of their first symptom.[16,17] While there are no population based studies on the epidemiology of DCM in SSA, hospital series reveal that DCM accounts for 20% of admissions to African hospitals for heart failure.[18] DCM has a 4 year mortality of 34% after onset of symptoms.[19]

Peripartum cardiomyopathy (PPCM): PPCM is a cardiomyopathy occurring between the commencement of the last month of pregnancy and the end of the fifth month in the postpartum

period, in women without preexisting symptoms, signs, or history of heart disease, and in the absence of any identifiable cause for the cardiac failure.[20-22] PPCM shares many common features with other forms of non-ischaemic cardiomyopathy.[18] The important distinction is that women with PPCM are younger, have a higher rate of spontaneous recovery of LV function, and have a better survival than patients with idiopathic DCM.[23,24] In a single centre prospective study of 100 South African patients with PPCM, 15% died, and only 23% recovered normal LV function after 12 months of treatment, despite optimal medical therapy with ACE inhibitors and β blockers.[25] In Nigeria, the contemporary mortality rate of PPCM is 42% over 25 years of follow-up.[26] Co-morbid HIV infection has not been shown to affect the outcome in PPCM patients.[27] PPCM has been shown to occur much more frequently in SSA than in developed countries. For instance, in the USA, the incidence of PPCM is 1 per 4000 births,[28] while in South Africa it is estimated to be 1 per 1000 births.[29] The highest incidence occurs in the Zaria province of Nigeria, and is reported to occur in 1 per 100 deliveries.[30] This observation of increased frequency in Nigeria may reflect volume overload as a result of ingestion of *kanwa*, a dried lake salt, while lying on heated mud beds twice daily for 40 days post-partum.[30] It is important to note, however, that this high incidence rate of PPCM was observed before modern diagnostic modalities like echocardiography had come into mainstream use.

Hypertrophic cardiomyopathy (HCM): HCM is an autosomal dominant disorder caused by mutation in genes encoding sarcomeric proteins, and has been shown to occur in 0.2% of 6680 unselected echocardiograms performed in Tanzania.[31] HCM was previously thought to be rare amongst Africans.[32] However, recent echocardiographic studies have dispelled that myth.[33,34] For example, in Ghana, HCM is the third commonest form of cardiomyopathy, after DCM and EMF.[35] In Ethiopia, HCM accounts for 34% of all cardiomyopathies diagnosed on echocardiography.[36]

1.3 The pathophysiology, natural history and outcomes in familial dilated cardiomyopathy

The first report of familial DCM was by Evans.[81] The first report of familial DCM in Africa described twin brothers in Uganda.[37] Brink *et al* subsequently described the interesting condition of 'hereditary dysrrhythmic congestive cardiomyopathy' in which prolonged QT interval is associated with ventricular arrhythmias and progressive dilatation of cardiac chambers.[38] DCM in sibling brothers has also been reported in South Africa.[39] Data from western populations demonstrate that 15-25% of all patients with DCM have a first degree relative that also show evidence of DCM suggesting familial transmission.[40] Furthermore, studies in South African patients with idiopathic DCM have revealed a common mitochondrial DNA variant that is associated with susceptibility to DCM in British and South African populations.[41] Maharaj and colleagues did HLA typing in 62 patients with DCM compared to 180 black normal controls to determine if immunogenetic factors could be involved in the pathogenesis of this condition.[42] In keeping with studies from western countries,[43,44] they demonstrated an association with HLA-DR1 and DRw10 antigens, implying that genetically determined immune response factors play a role in the pathogenesis of some individuals with DCM.[45] While no mutations have been found in the cardiac actin, skeletal actin, and AMPK genes[46] recently, mutations in the troponin T gene have been detected in a family with childhood onset DCM.[47]

In 1990, Geisterfer-Lowrance *et al* reported the first evidence of a gene defect underlying a form of intrinsic heart-muscle disease: they found that a mutation in the gene encoding the contractile protein ß-myosin heavy chain results in familial HCM.[48] Since then, mutations in several other genes, all encoding proteins of the myofibrillar apparatus, the fundamental contractile unit of the cardiomyocyte, have been found to cause familial HCM.[49,50] These proteins include sarcomeric proteins (ventricular myosin light chains 1 and 2, cardiac troponin I and T, α-tropomyosin, α-

cardiac actin) and an intra-sarcomeric cytoskeletal protein (myosin-binding protein C). Although the mechanisms by which mutations of the genes for these proteins lead to cardiac hypertrophy remain unclear, the elucidation of these gene defects provides important evidence of the primacy of impaired force generation in the pathogenesis of cardiac hypertrophy. Familial HCM is responsible for a small proportion of cases of cardiac hypertrophy. Nonetheless, it is of major clinical importance, since no specific therapy is available and it is the most common cause of sudden cardiac death (SCD) in young adolescents.[51] Thus, the discovery of gene defects that cause familial hypertrophic cardiomyopathy also heralds the exciting prospect of improved and early diagnosis and the potential for developing specific, mechanism-based therapies. [52]

In contrast to familial HCM, inherited forms of DCM account for only about 30 percent of cases of DCM and are a major cause of severe heart failure necessitating heart transplantation.[53] Most patients with idiopathic DCM initially present between the ages of 20 and 50 years, but the disorder may also affect children and the elderly.[51] The most common initial manifestation is heart failure (in 75 to 85 percent of patients), which is frequently advanced at presentation.[52] The chief morphologic feature is biventricular dilatation. Heart weight is increased, indicating hypertrophy, but maximal LV free-wall thickness and septal thickness are typically normal because of the abnormally dilated chambers. Functionally, the chief defect is impaired systolic performance, as opposed to the diastolic dysfunction characteristic of familial HCM.[53]

Although mutations in the gene for the extra-sarcomeric cytoskeletal protein dystrophin were identified some time ago as the cause of both muscular dystrophy (Duchenne type and Becker type) and X-linked DCM, the genetic causes of the inherited forms of DCM have, until recently, defied analysis.[54] In addition to defects in dystrophin (which may also cause myocarditis and DCM as a

result of cleavage by coxsackievirus B3),[55] other defects that might be responsible for DCM include those affecting the genes for α-cardiac actin[56] and those for two proteins — emerin and lamin — that, like the dystrophin-gene defect, can cause both muscular dystrophy and DCM.[57,58]

The identification of mutations in the gene for α-cardiac actin as a cause of DCM was surprising. Previously, α-cardiac actin had been regarded solely as a component of the sarcomere, and mutations in the gene for this protein, although distinct from those causing DCM, not unexpectedly were found to cause familial HCM.[50] More recent evidence suggests that α-cardiac actin may not only function as an important component of the sarcomere and, thus, be critically involved in force generation, but may also function as an intra-sarcomeric cytoskeletal protein that contributes to force transmission.[59] Hence, it appears that the same genes causing familial HCM may be responsible for familial DCM too.

1.4 The phenomenon of familial aggregation in peripartum cardiomyopathy

The familial aspects of dilated and hypertrophic forms of cardiomyopathy have been reviewed in the sections above. There are several studies that suggest significant familial clustering of PPCM. Pierce et al described in a series of 17 patients with PPCM, three of whom had a definite family history of PPCM (i.e. 18% prevalence of familial disease among the probands).[60] Massad et al published a case report of a woman with biopsy proven PPCM, whose sister had had a heart transplant for the same condition.[61] Pearl described a 24 year old white woman of Mexican origin dying of PPCM 17 days after delivery of her baby, who had a mother who also died at age 34 years of PPCM, a week after delivering her fifth child, as well as a sister who also died of congestive cardiac failure two days in the puerperium.[62] Voss et al described the case of PPCM, where the patient died of cardiac failure in the puerperium, as had her mother and two of her sisters.[63] Fett et al also reported a case of a mother and a daughter who both met the diagnostic criteria of PPCM.[64]

In a German study of PPCM patients, 15% were found to have probable familial cardiomyopathy, based on a positive family history of PPCM, DCM, SCD and arrhythmia.[65] Further, two separate studies have reported on common variants associated with PPCM.[66,67] A recent publication has reported on shared genetic predisposition between PPCM and DCM.[68]

1.5 Risk factors and outcomes in peripartum cardiomyopathy

Many authors have investigated risk factors for the development of PPCM. We have reviewed these previously.[23] These postulated risk factors include African ancestry, advanced maternal age, multiparity, poor socioeconomic status, multiple pregnancy, hypertension and prolonged tocolytic use. We have previously argued strongly for a stricter definition of PPCM that excludes pregnancy-related hypertensive disorders (including gestational proteinuric hypertension, eclampsia, pre-eclampsia, and post-partum hypertension).[23,69] In these hypertensive disorders of pregnancy, good LV recovery at six months is likely to be the norm, with minimal long term sequelae. This is contrasted with PPCM, where complete recovery of LV size and function occurs in about 30 to 50% of patients, with persistent LV dysfunction in the majority of survivors. Hence, it is our contention that hypertensive heart disorders of pregnancy should be viewed as a distinct clinical entity from PPCM, and not included in the clinical spectrum of PPCM, as is the current practice.[23,69] In PPCM specifically, non-systematic studies have revealed siblings of PPCM patients demonstrating LV dilatation and LV dysfunction. Even more striking, however, is that PPCM appears to share similar predisposing factors and clinical features with other forms of idiopathic (non-ischaemic) DCM; hence our proposition that they may be different manifestations of the same disease.[23] Further weight to support this view has been the shared genetic predisposition between PPCM and DCM, which is clearly not evident in those with HHFP.[68]

Several researchers have investigated outcomes in PPCM. Generally, the mortality in PPCM varies from 18-56% at 1 year.[70,71] In a prospective single centre South African study of 100 PCM patients, 15% died, while only 23% normalised their LV function at 1 year, despite optimal medical therapy.[25]

1.6 The Groote Schuur Hospital experience on cardiomyopathies

At Groote Schuur Hospital (GSH), we have abundant experience in managing patients with DCM, HCM, PPCM, ARVC, LVNC and RCM, with many end-stage patients receiving orthotopic heart transplantation (OHT). Much of this experience goes back to the time of the performance of the world's first heart transplant at GSH in 1967. We felt that it was timely to review our cumulative experience with these conditions, and to document the clinical phenotypes, clinical genetics and outcomes of our patients with these conditions, especially with the use of contemporary cardiovascular therapeutic agents.

1.7 Rationalé for the study

It is not clear whether the various phenotypic expressions of cardiomyopathy represent disparate clinical entities, or whether they are merely different forms of the same disease manifested differently in different circumstances. Furthermore, the familial aggregation of cardiomyopathy has not been studied systematically in an African setting. Moreover, the outcome of various cardiomyopathies in a contemporary African setting with current therapeutic modalities is unknown. Hence, we are unable to identify determinants of outcome as well as to prognosticate in the various cardiomyopathies. The proposed study envisages addressing these gaps in our knowledge, which will have implications for the management of patients with cardiomyopathy.

1.8 Hypothesis

We hypothesise that cardiomyopathies are a caused by familial/genetic and non-familial/non-genetic factors, and that disease onset, clinical and pathological features, disease progression and severity, and disease prognosis vary based on familial/genetic and non-familial/non-genetic aetiology.

1.9 Study aims

A number of studies have been conducted to test the above hypothesis. These studies aimed to investigate:

1. The frequency and clinical genetics of familial dilated cardiomyopathy (FDC);
2. The clinical characteristics and outcome of familial dilated cardiomyopathy compared to idiopathic dilated cardiomyopathy;
3. Risk factors and outcomes of PPCM;
4. The comparison of the clinical characteristics and outcome of PPCM versus hypertensive heart failure of pregnancy;
5. The genetics and outcomes of HCM;
6. Genotype-phenotype correlations of HCM in founder families examined by cardiac magnetic resonance imaging.

We investigated the above aims in a cohort of patients followed up over a 16 year period (1996-2009) at GSH. In particular, we were interested in exploring the demographic parameters of this group, survival and other outcome parameters, as well as the determinants of survival in specific cohorts. We also studied family pedigrees in order to be able to characterise the clinical genetics of familial DCM in this cohort.

Chapter 2

Methods

A detailed description of the Methods is included in each of the chapters that follow. Briefly, the Methods are described below.

2.1 Study design

The sub-studies included in this thesis were designed either as retrospective analysis of information obtained from hospital records of patients previously seen at the GSH Cardiac Clinic or cross-sectional studies of CMR imaging in probands and first degree relatives of patients with HCM from Tygerberg Hospital scanned at GSH.

2.2 Subject selection

The inclusion criteria for the study was that patients must have been diagnosed with cardiomyopathy, based on existing clinical case definitions. Patients were be identified from the GSH Cardiac Clinic and cardiac genetic database records.

2.3 Measurements

A data collection database that incorporated patient demographic details, medical history and co-morbidity, as well as details of imaging, laboratory and invasive investigations and management was utilised.

2.4 Statistical Analysis

Simple descriptive statistics were used both for data interpretation and to draw inferences about the population of patients that were studied. SPSS and STATISTICA were used for univariable and multivariable regression analysis and for performing correlations on risk factors and outcomes. Kaplan-Meier plots were used to demonstrate survival.

2.5 Ethical considerations

The study was carried out in the spirit of the Helsinki declaration. The aims, design and proposed conduct of the study reflect paragraph six of the Helsinki declaration, which reads: "The primary purpose of research involving human subjects is to improve prophylactic, diagnostic and therapeutic procedures and the understanding of the aetiology and pathogenesis of disease." This study was formulated in compliance with principles delineated in the Helsinki declaration (last modified in 2013).

All research procedures were reviewed and approved by the institutional review board of the University of Cape Town Faculty of Health Human Research Ethics Committee. Initially, we

obtained approval from the University of Cape Town Research Ethics Committee to perform a clinical and genetic study of all cardiomyopathies (UCT REC 197/1996). Subsequently, we were granted permission to perform a familial aggregation study of PPCM (UCT REC 020/2007). Furthermore, we also had approval for the study of HCM and other cardiomyopathies using CMR – the Cape Cardiac MRI initiative (UCT REC 307/2004). All subjects gave informed consent for their participation in these studies.

Chapter 3

Frequency and clinical genetics of familial dilated cardiomyopathy in Cape Town: implications for the evaluation of patients with unexplained cardiomyopathy

3.1 Introduction

DCM is defined by the presence of LV systolic dysfunction and dilatation in the absence of abnormal loading conditions (hypertension, valve disease) or coronary artery, pericardial or congenital heart disease sufficient to cause global systolic impairment.[5] Right ventricular dilation and dysfunction may be present but are not necessary for the diagnosis.[72] DCM is an important cause of heart failure, as well as a common indication for heart transplantation.[52] The clinical and morphological diversity of DCM reflect the broad spectrum of distinct underlying molecular and environmental causes.[53] The prevalence of DCM is estimated to be 36.5 per 100, 000 in a United States population.[73] There are, however, no population-based studies of the epidemiology of DCM in sub-Saharan Africa.[12] Most cases of DCM are thought to be sporadic or acquired.[74] However, in many cases the disease is inherited and is termed familial DCM, which may account for up 25-50% of DCM in Western populations.[75-77]

Familial DCM is principally caused by genetic mutations in genes that encode cytoskeletal, nuclear and sarcomeric proteins in the cardiac myocyte.[78] In addition, modifying genes, lifestyle and additional factors are reported to influence onset of disease, disease progression, and prognosis.[79]

Pedigree analysis in familial DCM is most consistent with autosomal dominant inheritance with variable penetrance.[80]

To the best of our knowledge, there is no information on the frequency and clinical genetics of familial DCM in Africa. The aims of this sub-study were three fold: first, to describe the frequency of familial DCM in a series of patients who were diagnosed with DCM and followed-up in a cardiomyopathy clinic at the Cardiac Clinic at GSH in Cape Town from 01 February 1996 to 31 December 2009; second, to document, through pedigree analysis, the likely modes of inheritance of familial DCM in the Cape Town cohort; and finally, to address the implications of the findings for the clinical evaluation of patients with unexplained DCM.

3.2 Methods

3.2.1 Study design

The study was designed as a prospective hospital-based study of the frequency and clinical genetics of familial DCM in a cohort of DCM patients referred to the cardiomyopathy clinic in the Cardiac Clinic, GSH, Cape Town, South Africa. The frequency of familial DCM was based on a detailed family history in all individuals and family screening of first-degree relatives of probands with a positive history for confirmation of familial disease. Familial DCM was defined as the presence of DCM in at least one first degree relative of the proband and/or the presence of sudden unexplained death under the age of 35 years in a first degree relative.

3.2.2 Study population

We reviewed the medical records of all patients evaluated for a cause of cardiomyopathy from 01 February 1996 and 31 December 2009. The patients in this study were seen in a dedicated cardiomyopathy clinic. All patients included in the study were those with DCM, as evidenced by clinical signs and symptoms of heart failure associated with a LV dilatation and a LV ejection fraction less than 50% on echocardiography or cardiac catheterisation. Patients with any of the following conditions were excluded: rheumatic or other intrinsic valvular heart disease, coronary artery disease (CAD), pericardial disease, chronic hypertension, congenital heart disease, myocarditis and any other systemic disease with cardiovascular sequelae such as diabetes, drug-induced cardiomyopathy, haemochromatosis, neuromuscular disease, infantile endocardial fibroelastosis, EMF, amyloidosis, or myocarditis documented on endomyocardial biopsy (EMB) or associated with proven viral infection. Patients with alcohol ingestion were included, since such persons may have a genetic predisposition to DCM. Furthermore, their exclusion could spuriously inflate the proportion of patients with familial DCM since a diagnosis of alcohol-induced cardiomyopathy might be made more readily in absence of a family history of DCM. For similar reasons, patients with PPCM were included. However, patients with HCM, RCM and tachycardia-related cardiomyopathies were excluded.

3.2.3 Clinical genetics

A two to five generation family pedigree was constructed for every patient with DCM. If a subject with DCM had any first-degree relative that was also affected with DCM, those first-degree relatives were invited for screening for DCM by history, physical examination, electrocardiogram, echocardiography and cardiac catheterisation where indicated. For deceased relatives, medical records were reviewed when available. Relatives with other cardiac disorders that could account for LV dysfunction were excluded from the analysis. Pedigree analysis was performed for all affected

families for likely pattern of inheritance of familial DCM. Pedigrees were constructed using the Cyrillic 2.1 software programme (http://www.cyrillicsoftware.com/).

3.2.4 Statistical analysis

Results of quantitative variables are given as mean ± standard deviation (SD). Categorical variables are represented as number and percentage. Pearson's chi-square or Fisher's exact test were used to compare the relative frequency of characteristics between individuals. All P values are two-sided; and P values < 0.05 are considered to indicate statistical significance.

3.2.5 Ethical considerations

The study was approved by the University of Cape Town Faculty of Health Sciences Human Research Ethics Committee (REC Ref No. 197/96). All participants gave written informed consent to participate in the study. All eligible patients were asked for permission to invite their first-degree relatives (parents, siblings and children) to participate in the study.

3.3 Results

3.3.1 Frequency of familial DCM

109 unrelated cases with DCM were studied, 29 of whom had familial disease; hence, the frequency of familial DCM was 26.6%. The 29 cases were derived from all the populations of Cape Town, and reflected the referral base of Groote Schuur Hospital; 15 (48.3%) were coloured/mixed ancestry African, 10 (34.5%) were black African, 4 (13.8%) were white African, and 1 (3.4%) was of Indian descent. A total of 40 individuals with familial DCM, belonging to the 29 different families with familial DCM, were identified through family screening.

3.3.2 Characteristics of patients with familial DCM

The 29 unrelated cases with familial DCM had a mean (±SD) age at diagnosis of 28.01 ± 15.3 years with a preponderance of males (n 21, 72.5%); these characteristics were comparable to the total of 40 individuals with familial DCM from the 29 families who were studied. The age of individuals with familial DCM was significantly lower than that of patients with non-familial DCM, who had a mean age of 39.1 ± 12.6 at the time of diagnosis (p=0.001) (Table 3.1).

Table 3.1. Clinical characteristics of patients with familial and non-familial DCM

Characteristic	Index cases with familial DCM (n = 29)	Non-familial DCM (n = 80)	P value
Age in years at diagnosis (mean ±SD)	28.01 ± 15.33	39.10 ± 12.6	0.001
Gender (%)			
Males	21 (72.4)	48 (60)	
Females	8 (27.6)	32 (40)	0.260
Ethnicity (%)			
Black African	10 (34.5)	35 (43.8)	
White African	4 (13.8)	13 (16.3)	
Indian ancestry	1 (3.4)	0 (0)	
Coloured/Mixed ancestry	14 (48.3)	32 (40)	0.602
Home language (%)			
isiXhosa	8 (27.6)	32 (40)	
English	6 (20.7)	11 (13.8)	
Afrikaans	14 (48.3)	35 (43.8)	
Other	1 (3.4)	2 (2.5)	0.417
NYHA functional class (%)			
Class I and II	8 (27.6)	19 (23.8)	
Class III and IV	21 (72.4)	61 (76.3)	0.110
HR at initial presentation (bpm) ± SD	96.02 ± 17.98	94.89 ± 19.53	0.496
Systolic BP (mmHg) ± SD	102.37 ± 15.05	101.78 ± 18.11	0.397
LBBB (%)	5 (17.2)	29 (36.3)	0.123
LVEDD (cm) ± SD	6.28 ± 1.19	6.84 ± 1.37	0.026
LVEF (%)± SD	28.10 ± 10.98	24.68 ± 11.50	0.048

DCM = dilated cardiomyopathy; NYHA = New York Heart Association; HR = heart rate; BP = blood pressure; LBBB = left bundle branch block; LVEDD = left ventricular end diastolic dimension; LVEF = left ventricular ejection fraction.

There was no difference in degree of effort intolerance measured by the New York Heart Association (NYHA) Functional Class at presentation between the index cases with familial and non-familial DCM (Table 3.1). The patients with familial and non-familial DCM also had similar heart rate and mean systolic blood pressure at presentation (i.e., 95.6 ± 23.3 beats per minute versus 94.9 ± 19.5 bpm [p=0.496] and 100.1 ± 14.9 mmHg versus 101.8 ± 18.1 mmHg [p=0.397; respectively). Whilst the frequency of electrocardiographic left bundle branch block was lower in the familial cases (17.2%) compared to the non-familial cases (36.3%), the difference was not statistically significant (p=0.123). However, familial DCM cases had significantly less ventricular dilation than non-familial cases; the left ventricular end diastolic dimension (LVEDD) was 6.2 ± 1.1 cm in familial DCM patients versus 6.8 ±1.4 cm in non-familial DCM patients, p=0.001 (Table 3.1).

3.3.3 Clinical genetics of familial DCM

Table 3.2 contains a summary of the pedigree analysis of the families with DCM that were studied. Pedigree analysis of the 29 families was consistent with autosomal dominant (AD) inheritance in 21 (72.4%) families. An autosomal recessive (AR) inheritance pattern was observed in 5 (17.2%) families. X-linked recessive (X-LR) inheritance was seen in 3 (10.4%) of the familial DCM families. The examples of family pedigrees with AD, AR, and X-LR inheritance are depicted in Figure 3.1, and the full list of pedigrees is available as supplementary information online at http://www.medicine.uct.ac.za/.

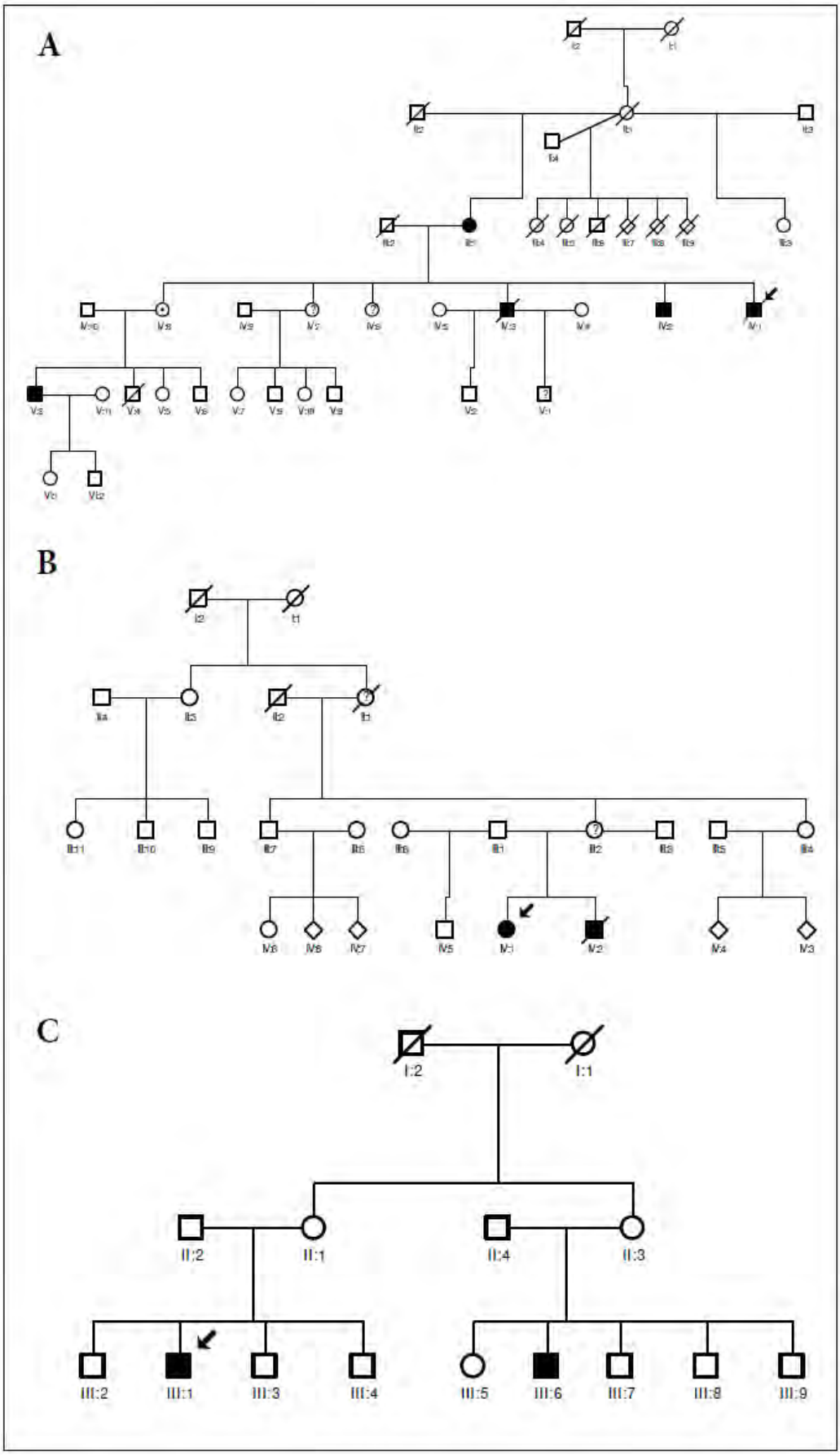
Table 3.2. Clinical genetics of the 29 families with familial dilated cardiomyopathy

Mode of inheritance	Number (%)
Autosomal dominant	21 (72.4)
Autosomal recessive	5 (17.2)
X-linked recessive	3 (10.4)
Number of affected 1st-degree relatives per family (mean ±SD)	2.16 ± 1.14
Number of 1st-degree relatives at risk per family (mean ±SD)	3.45 ± 2.23
Number of families with a family history of SCD (%)	1 (3.4)
Number families with affected 2nd degree relatives (%)	9 (31)

SCD, sudden cardiac death under the age of 35 years

The number of affected first-degree relatives per family was 2.16 ± 1.14. In these 29 families with DCM, the number of first-degree relatives at risk was 3.45 ± 2.23 per family. One family (3.4%) had a clear history of a member with sudden cardiac death (SCD) before the age of 35 years. Of the 29 families included in the analysis, 9 (31.0%) had second degree relatives who were affected.

It is noteworthy that two of the 29 unrelated patients (7%) with familial DCM had at least one relative who was diagnosed with PPCM, confirming that some cases of PPCM are part of the spectrum of familial DCM.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III:7
III:8
III:9

Figure 3.1. Pedigrees and mode of inheritance

Pedigree analysis of the 29 families was consistent with autosomal dominant inheritance (e.g. panel A) in 72.4% (N=21); autosomal recessive inheritance (e.g. panel B) in 17.2% (N=5) and X-linked recessive inheritance (e.g. panel C) in 10.4% (N=3). Filled symbol = phenotypically affected subject; empty symbol = unaffected subject; circle = woman; square = man; diamond = gender unknown; arrow = proband; bar crossing the symbol = deceased. All the pedigrees are available as supplementary material online at http://www.medicine.uct.ac.za

3.4 Discussion

In 1949, Evans was the first to report on familial cases of cardiomyopathy,[81] and subsequently there have been many publications on familial DCM. This study extends the observations that have been made in Western populations to Africa. We show that familial DCM occurs in 26.6% of cases of DCM, that it is a disease of young males with an autosomal dominant pattern of inheritance in the majority of families. We also confirm that some cases of PPCM, which occurred in the context of familial DCM in two families, are part of the spectrum of familial DCM.

The finding of familial disease in a quarter of DCM patients in this study is in line with several other reports that have found inherited disease to constitute 20 to 50% of DCM patients.[73-75,82,83] A prospective echocardiographic study that screened first-degree relatives of patients with DCM estimated the rate of familial DCM at 20.3%,[75] as 315 relatives of 59 index patients with DCM underwent screening with echocardiography, including coronary angiography for those older than 40 years in order to exclude CAD. DCM was found in 18 relatives (20% of index patients), whereas only 5% had been suspected of having familial disease based on family history alone. In another prospective study of 56 probands with DCM, the definite familial DCM rate was estimated at 25%, where the diagnosis of familial diseased was based on first-degree relatives with a diagnosis of DCM on cardiac catheterisation or on autopsy, or a first-degree relative with both an echocardiographic LV end-diastolic dimension (LVEDD) greater than two standard deviations above the mean and a LV ejection fraction (LVEF) below 50%.[19] When LV enlargement was taken

as a clinical indicator of DCM, two other studies found the frequency of familial DCM to range from 29% to 48%.[84,85]

In our study, over 70% of patients with familial DCM were male. This male preponderance in DCM has been previously described.[86,87] In SSA, DCM has been shown in hospital-based studies to occur twice as commonly in men as in women.[88] Moreover, a study of 637 DCM patients, including 130 patients with familial DCM, found that male patients constitute approximately 70% of both non-familial DCM and familial DCM cohorts studied.[89]

The finding of a younger age of onset of familial DCM compared to non-familial disease is also consistent with other studies and compatible with a genetic cause. Moretti and colleagues have found familial DCM patients to be younger than non-familial cases (40 years vs. 48 years).[89] One of the earlier systematic studies of familial DCM by Michels *et al* also found the disease to occur at a younger age (mean age of onset of 32 years) when compared to patients with idiopathic DCM.[90] A different study comparing familial DCM and non-familial DCM found that only a younger age of onset was predictive of familial disease; no other clinical or morphological features were useful in distinguishing the two entities from each other.[85] Likewise, Grünig and co-investigators found that 156 of 445 DCM patients confirmed to have familial DCM, were much younger than those without familial disease.[91] Early-onset CVD is likely to be influenced by genetic factors more than late-onset disease.[92]

We found that at least 70% of familial DCM families demonstrated an autosomal dominant (AD) pattern of inheritance. This observation is supported by many prior publications. Familial DCM has been reported most commonly with AD inheritance in up to 90% of cases.[77,85,92,93] The genetic and

clinical heterogeneity observed is in keeping with causation by multiple genes, with gene-environment interactions altering expression of disease.[85] To date, over 25 mutations have been described in autosomal genes that are causative of familial DCM.[77,92] Some of the commonly mutated genes causing AD type of familial DCM include lamin A/C, beta-myosin heavy chain, alpha-myosin heavy chain, actin, alpha-actinin-2, metavinculin, desmin, sarcoglycan, troponin T, alpha-tropomyosin, titin and phospholamban.[77,93] There are also genetic polymorphisms that are associated with an increased risk of developing DCM in some populations.[41,94]

An AR pattern of inheritance may account for up to 10% of familial DCM, and is commoner in certain ethnic groups and in cases of infantile DCM.[95] Unlike many other studies, we found a slightly higher rate of AR inheritance in our study: 17.2%. A mutation in cardiac troponin I has been shown to cause AR DCM in one family.[96]

X-linked familial DCM has been reported in 5-10% of cases.[87] X-linked types of familial DCM usually result from mutations in the dystrophin gene, which are commonly associated with skeletal muscle weakness and elevated levels of creatinine kinase.[77,97,98] In some cases, DCM has been noted to be the only presenting feature of patients with Becker muscular dystrophy or in female carriers.[99] Furthermore, Becker muscular dystrophy and DCM have been seen in the same family, suggesting that it may be difficult to draw conclusions about phenotype from genotype.[100] Sporadic dystrophin mutations are also identified in sporadic forms of X-linked DCM.[101]

The determination of inheritance patterns by clinical genetic analysis of familial DCM is not straight-forward. Use of family history and construction of a three- or four-generation family pedigree may not be sensitive enough, when used alone for screening. Often, the phenotypes of affected individuals are not sufficiently specific for familial DCM. Furthermore, familial DCM

demonstrates incomplete penetrance, age-dependent disease expression and variable expression.[85] Among individuals carrying the same gene mutation, there may be wide variability in phenotypic effects and disease severity both within and between families. Within the same family, the phenotype may range from subtle or no symptoms to development of arrhythmia, heart failure, sudden cardiac death, complication with stroke or need for cardiac transplantation.[102]

The observations in this study have major implications for clinical practice. Inherited forms of cardiomyopathy are frequently responsible for heart failure that is otherwise unexplained, and often labeled as idiopathic cardiomyopathy before full evaluation has been conducted. Evaluation of familial cardiomyopathy should include not only the individual patient, but also the pattern of inheritance within the family and assessment for the presence of syndromic features. The last 10 years have seen remarkable advances in genetics. Improvements in technology have lowered costs, such that clinical use of molecular genetic testing is rapidly expanding in South Africa[103] and other parts of the world. Genetic counseling about the potential risks and benefits of genetic testing is an essential part of the clinical care of individuals and families with inherited heart disease. However, the likelihood of finding a responsible gene mutation varies among the different types of inherited cardiomyopathy. Both hypertrophic and right ventricular forms of cardiomyopathy have a relatively high likelihood of finding a responsible gene mutation when molecular genetic testing is properly applied. By contrast, the identification of pathogenic mutations in familial DCM is more challenging because of extreme genetic heterogeneity. Nevertheless, the clinical screening of family members who are at risk for an inherited form of cardiomyopathy leads to earlier identification, earlier treatment, and improved outcomes with or without molecular genetic testing.

This sub-study has several limitations. First, a relatively small number of families are included. Second, all these patients were seen in a tertiary referral centre, where there may be referral bias, which may overestimate the true frequency of familial DCM. However, on the other hand, the number of familial DCM cases in our cohort may have been underestimated for at least two reasons: (1) A varying spectrum of cardiac abnormalities may be present in asymptomatic relatives of patients with familial DCM, hence, familial and sporadic types of the disease are not easily distinguishable by family history and clinical screening of first-degree relatives in the absence of a 'gold standard' for diagnosis; (2) in our study, the diagnosis of familial DCM was based on family history, and screening of first degree relatives of individuals with a positive family history to confirm the presence of familial DCM. It is likely therefore that we report the minimum frequency of familial DCM, and that the screening of the individuals without a family history is likely to yield a higher prevalence of familial disease.

3.5 Conclusions

We have shown that familial DCM is common in African patients with unexplained DCM, occurring in a quarter of DCM patients studied. We also found that familial DCM has a predilection for males as well as an early age of onset, with the mean age at diagnosis being 28 years. Over 70% of the affected families demonstrated an autosomal dominant pattern of inheritance. These findings have major implications for the clinical evaluation of patients with unexplained cardiomyopathy (including those with PPCM), in whom family screening of first degree relatives for cardiomyopathy is indicated.

Chapter 4

Clinical characteristics and outcomes of familial and idiopathic dilated cardiomyopathy in Cape Town: a comparative study of 120 cases followed up over 14 years

4.1 Introduction

Cardiomyopathies are primary disorders of heart muscle associated with LV dysfunction and development of heart failure, in which the heart muscle is structurally and functionally abnormal in the absence of significant coronary artery disease, hypertension, valvular disease, pericardial disease or congenital heart disease sufficient to explain the observed myocardial abnormality.[5] In SSA, DCM accounts for 10% to 17% of all cardiac conditions encountered at autopsy, and for 17% to 48% of patients hospitalised for heart failure.[12,33]

The Heart of Soweto study (a contemporary study examining the characteristics and burden of CVD in urban Africa) indicates that heart failure is the commonest cardiovascular diagnosis, with moderate-to-severe LV dysfunction present in 53% of cases, and 68% of those with heart failure diagnosed with DCM or hypertensive heart disease.[104] While DCM occurs at any age, it is common in the third and fourth decades of life, with men affected twice as commonly as women. Two thirds of patients with DCM, especially those who are more than 55 years of age, have persistently low arterial pressure, ventricular arrhythmias, and/or atrioventricular valve incompetence and die within 5 years of their first symptom.[17]

To the best of our knowledge, with the exception of PPCM,[25] there are no studies of the outcome of DCM in a contemporary African setting. While many cases of DCM are thought to be sporadic or acquired, in up to 50% of patients, the disease is inherited and is termed familial DCM.[105] Familial DCM is principally caused by mutations in genes encoding cytoskeletal, nuclear and sarcomeric proteins in the cardiac myocyte, often presenting with incomplete penetrance, variable expression and significant locus and allelic heterogeneity.[106] Modifying genes, lifestyle and additional factors are reported to influence onset of disease, disease progression, and prognosis in familial DCM. It is of clinical interest to know whether the clinical characteristics and treatment modalities influence outcome in patients with familial DCM, and whether there are differences in clinical characteristics and outcome between those with familial disease compared to idiopathic cases of DCM.

The purpose of this sub-study was to compare the clinical characteristics and outcome of patients with familial and idiopathic DCM in a contemporary African setting with access to a full range of proven medical and surgical interventions for heart failure, such as cardiac resynchronisation therapy (CRT) and OHT, which are available at GSH.[107]

4.2 Methods

4.2.1 Study design

The sub-study was designed as a prospective hospital-based study of the clinical characteristics and outcomes of familial and DCM at GSH, Cape Town, South Africa. We reviewed the medical records of all patients diagnosed with idiopathic DCM, evaluated at GSH Cardiac Clinic from February 1, 1996 and December 31, 2009.

4.2.2 Study population

The patients in this study were seen in a dedicated cardiomyopathy clinic. All patients included in the study were diagnosed with using established criteria for DCM, based on evidenced by clinical heart failure associated with a LV dilatation and a LVEF less than 50% on echocardiography or cardiac catheterisation. Familial DCM was defined as the finding of a first-degree relative with DCM in a patient with unexplained DCM. The selection of the patients for inclusion in this study has been described in detail before. Patients with valvular heart disease, CAD, pericardial disease, chronic hypertension, congenital heart disease, myocarditis and any other systemic disease with cardiovascular sequelae were excluded.

4.2.3 Data collection

All patients had comprehensive clinical assessment. Information collected at the time of physical examination included history of medical co-morbidity, medications used, history of alcohol and tobacco use. Important clinical parameters including the pulse, blood pressure, oedema, the height of the jugular venous pressure, presence of murmurs and crepitations were recorded. Clinical assessment was complemented by chest radiography, electrocardiography, and detailed two-dimensional and Doppler colour-flow echocardiography. Normal values for echocardiographic measurement were based on age and body-surface area. Cardiac catheterisation was performed, when appropriate, and tissue obtained at biopsy of right ventricular endomyocardium was examined by light microscopy, immunohistochemistry and electron microscopy by a pathologist with no knowledge of the family history of any patient.

4.2.4 Statistical analysis

Results of quantitative variables are given as mean ± SD. Categorical variables are represented as number and percentage. Pearson's chi-square or Fisher's exact test was used to compare the relative

frequency of characteristics between individuals. All P values were two-sided; and a P value <0.05 was considered significant.

Kaplan-Meier survival curves were constructed using the product-limit method, and were compared using the log-rank test. Kaplan-Meier curves were utilised to visualise the survival experience between familial and idiopathic DCM patients, and the focus is on mortality, rather than time to death, justifying the choice of Cox's logistic regression rather than the Cox proportional hazards model. Age-, gender and race-adjusted survival curves for the general South African population were derived and compared with the Kaplan-Meier survival rates for the patients with idiopathic or familial DCM using the chi-square test. Kaplan-Meier survival curves were constructed using STATA version 9.0 (STATA Corporation, College Station, TX, USA). Univariate and multivariate logistical regression analysis was used to determine predictors of death.

4.2.5 Ethical considerations

The study was approved by the University of Cape Town Faculty of Health Sciences Human Research Ethics Committee (REC Ref No. 197/96). All participants gave written informed consent to participate in the study. All eligible patients were asked for permission to invite their first-degree relatives (parents, siblings and children) to participate in the study.

4.3 Results

4.3.1 Clinical characteristics at presentation

A total of 120 patients were studied, comprising 80 idiopathic DCM patients and 40 familial cases from 29 families. The familial DCM patients had a younger age at diagnosis compared to the

idiopathic DCM patients, with mean ages of 25.6 ± 15.1 years and 39.1 ± 12.6 years (p=0.001), respectively, as reported elsewhere.

The clinical symptoms and signs of patients with familial DCM were similar to those of patients with idiopathic DCM (Table 4.1). Electrocardiographic findings were similar between the two groups, with the exception of T wave inversion, which was found more commonly in familial patients compared to idiopathic DCM patients: 35 (87.5%) vs. 55 (68.8%), respectively (p=0.014), and pathological Q wave abnormalities which were found more commonly in idiopathic DCM (32.5%) than in familial cases (12.5%) (p=0.028). Patients with idiopathic DCM were more likely to have radiographic cardiomegaly, as found in 75/80 (93.8%) of idiopathic DCM patients compared to 28/40 (70%) of familial DCM patients (p=0.020). The radiographic findings were corroborated by echocardiography, where idiopathic DCM patients were found to have significantly larger LV end-diastolic dimensions (LVEDD) (p=0.001), lower LVEF (p=0.026) and fractional shortening (FS) (p=0.048) than the familial DCM cases.

At cardiac catheterisation, the only difference noted was in the LVEF, which was found to be lower in idiopathic DCM patients compared to familial DCM patients (26.2 ± 9.7 vs. 31.07 ± 15.63 %, p=0.032). There were no differences in the histological characteristics on light microscopy between the two groups. There were no significant differences between familial and idiopathic DCM patients in relation to risk factors for CVD, lung disease, HIV infection and stroke.

Table 4.1. Clinical, radiographic, electrocardiographic and echocardiographic features at presentation

Clinical characteristics	***Idiopathic DCM (N = 80)***	***Familial DCM (N = 40)***	***P-value***
NYHA FC (%)			
Class 1 and 2	19 (23.8)	11 (27.5)	
Class 3 and 4	61 (76.3)	29 (72.5)	0.110
Dyspnoea (%)	79 (98.8)	36 (90)	0.295
Fatigue (%)	79 (98.8)	40 (100)	0.596
Angina/chest pain (%)	9 (11.3)	4 (10)	0.489
Palpitations (%)	19 (23.8)	10 (25)	0.513
Heart rate (%)	94.89 ± 19.53	95.61 ± 28.32	0.496
Blood pressure [systolic] (%)	101.78 ± 18.11	100.10 ± 14.94	0.397
Blood pressure [diastolic] (%)	64.37 ± 12.82	70.29 ± 13.45	0.289
Pedal oedema (%)	59 (73.8)	29 (72.5)	0.583
JVP height [cm] (%)			
Below 3cm	23 (28.7)	11 (27.5)	
3cm to angle of jaw	28 (35.0)	11 (27.5)	
Above angle of the jaw	29 (36.3)	18 (45)	0.337
Murmur (%)			
No murmur	18 (22.5)	12 (30)	
MR	35 (43.8)	13 (32.5)	
MR + TR	24 (30)	14 (35)	
ESM	3 (3.8)	1 (2.4)	0.220
S3 Gallop (%)	56 (70)	27 (67.5)	0.695
Basal crepitations (%)	39 (48.8)	24 (60)	0.165
Radiographic features			
Cardiothoracic ratio >50% (%)	75 (93.8)	28 (70)	0.020
Radiographic pulmonary oedema (%)	43 (53.8)	24 (60)	0.412
Electrocardiographic features			
Heart rate ± SD	92.85 ± 19.53	96.58 ± 23.02	0.498
Sinus rhythm (%)	60 (75)	26 (65)	0.218
QRS abnormalities present (%)	36 (45)	14 (35)	0.399
Increased voltage (%)	40 (50)	22 (55)	0.583
Presence of pathological Q waves (%)	26 (32.5)	5 (12.5)	0.028
Left atrial hypertrophy (%)	19 (23.8)	8 (20)	0.318
Left bundle brunch block morphology (%)	29 (36.3)	7 (17.5)	0.123
Right bundle brunch block morphology (%)	2 (2.5)	0 (0)	0.518
Left ventricular hypertrophy (%)	28 (35)	12 (30)	0.243
Right ventricular hypertrophy (%)	1 (1.3)	0 (0)	0.721

PR prolongation present (%)	2 (2.5)	0 (0)	0.311
T wave inversion (%)	55 (68.8)	35 (87.5)	0.014
Arrhythmia present (%) No arrhythmia Atrial fibrillation Atrial flutter Ventricular tachycardia Paced rhythm	 60 (75) 16 (20) 0 (0) 1 (1.3) 3 (3.8)	 26 (65) 10 (25) 1 (2.5) 2 (5) 1 (2.5)	 0.179
QRS duration (ms)	112.07 ± 11.93	109.98 ± 12.31	0.107
Echocardiographic features			
Interventricular septal thickness (systole) ± SD	1.04 ± 0.36	1.15 ± 0.39	0.469
Interventricular septal thickness (diastole) ± SD	0.92 ± 0.30	1.08 ± 0.36	0.389
Left ventricular posterior free wall thickness (systole) ± SD	1.10 ± 0.41	1.18 ± 0.42	0.292
Left ventricular posterior free wall thickness (diastole) ± SD	0.92 ± 0.32	0.96 ± 0.30	0.593
Left ventricular dimension (systole) ± SD	5.54 ± 1.31	5.42 ± 2.13	0.602
Left ventricular dimension (diastole) ± SD	6.84 ± 1.37	6.21 ± 1.13	0.001
Left ventricular ejection fraction ± SD	24.68 ± 11.50	28.01 ± 10.51	0.026
Left ventricular fractional shortening ± SD	12.10 ± 5.99	14.81 ± 7.86	0.048

ESM, ejection systolic murmur; JVP, jugular venous pressure; MR, mitral regurgitation; NYHA FC, New York Heart Association functional class; TR, tricuspid regurgitation

4.3.2 Medical therapy and outcome at last follow-up visit

Familial and idiopathic DCM patients were on similar treatments. However, beta-blockers (p=0.007) and digoxin (0.028) were more commonly prescribed for idiopathic DCM patients (Table 4.2). There were no differences in mortality, clinical features, cardiovascular complications or utilisation of OHT between idiopathic DCM and familial DCM patients at last follow-up (Table 4.3).

Table 4.2. Medical therapy at follow-up

Medical therapy at last follow-up visit	*Idiopathic DCM (N = 80)*	*Familial DCM (N = 40)*	*P-value*
Furosemide (%)	78 (97.5)	37 (92.5)	0.116
ACE-I* or ARB** (%)	80 (100)	39 (97.5)	0.466
Beta-blocker (%)	76 (95)	30 (75)	0.007
Digoxin (%)	70 (87.5)	26 (65)	0.028
Spironolactone (%)	62 (78.5)	25 (62.5)	0.072
Calcium channel blocker (%)	0 (0)	3 (7.5)	0.169
Warfarin (%)	22 (27.5)	12 (30)	0.445

*ACE-I, angiotensin converting enzyme inhibitor; **ARB, angiotensin receptor blocker

Table 4.3. Complications, treatment and outcome data

Outcome data	*Idiopathic DCM (N = 80)*	*Familial DCM (N = 40)*	*P-value*
Median duration of follow-up in years (IQR)	5.58 (1.42 – 10.41)	4.58 (1.33 – 14.8)	0.063
Death at the end of the median follow-up period (%)	32 (40)	16 (40)	0.595
Chronic heart failure (%)	63 (78.8)	32 (80)	0.515
Intra-cardiac clot (%)	3 (3.8)	2 (5)	0.496
Embolic phenomena (%)	2 (2.5)	1 (2.5)	0.505
Pulmonary hypertension (%)	0 (0)	1 (2.5)	0.466
Implantable cardioverter defibrillator insertion (%)	1 (1.3)	1 (2.5)	0.453
Permanent pacemaker insertion (%)	2 (2.5)	1 (2.5)	0.513
Cardiac resynchronisation therapy (%)	2 (2.5)	3 (7.5)	0.108
Orthotopic heart transplantation (%)	15 (18.8)	11 (27.5)	0.116
NYHA FC at last visit (%)			
Class 1 and 2	50 (62.5)	17 (42.5)	
Class 3 and 4	30 (37.5)	23 (57.5)	0.205

NYHA FC, New York Heart Association functional class

On univariate logistic regression analysis, factors that increased the likelihood of mortality in both familial and idiopathic cases were an increased LVEDD (p=0.038) and LVESD (p=0.049), an elevated JVP (above the angle of the jaw) at the initial visit (p=0.028), pulmonary hypertension (p=0.025), the lack of OHT (p=0.004) and symptomatic heart failure with NYHA functional class III and IV symptoms at the last visit (p=0.017), as shown in Table 4.4a. On multivariate analysis,

the significant predictors of mortality include lack of OHT (OR 4.7 [1.3 – 72.6], p=0.026) and NYHA class III and IV symptoms at the last visit (OR 3.8 [1.3 – 48.5], p<0.001).

The use of digoxin in idiopathic DCM patients was associated with increased mortality (OR 1.6 [1.0 – 3.9], p=0.037), but not in familial DCM patients (Table 4.4b).

Table 4.4a. Logistical regression analysis for predictors of mortality in both familial DCM and idiopathic DCM

Characteristic	***Univariate analysis***		***Multivariate analysis***	
	Odds ratio (95% CI)	**P-value**	**Odds ratio (95% CI)**	**P-value**
LVESD	1.764 (1.133 – 2.747)	**0.038**	0.955 (0.770 – 1.184)	0.674
LVEDD	1.719 (1.129 – 2.618)	**0.049**	1.117 (0.889 – 1.403)	0.341
JVP (>AOJ vs. <AOJ)	5.600 (1.202 – 26.095)	**0.028**	1.808 (0.256 – 12.777)	0.553
Pulmonary hypertension (yes vs. no)	3.586 (1.086 – 13.660)	**0.025**	1.401 (0.941 – 2.360)	0.646
Orthotopic heart transplantation (no vs. yes)	8.053 (0.954 – 67.970)	**0.004**	4.717 (1.306 – 72.600)	**0.026**
NYHA FC at last visit (class 3 or 4 vs. class 1 or 2)	12.473 (1.576 – 98.703)	**0.017**	3.848 (1.305 – 48.469)	**<0.001**

LVESD, left ventricular end systolic dimension; LVEDD, left ventricular end diastolic dimension; JVP, jugular venous pressure; NYHA FC, New York Heart Association Functional Class

Table 4.4b. Logistical regression analysis for predictors of mortality in idiopathic DCM

Characteristic	***Univariate analysis***		***Multivariate analysis***	
	Odds ratio (95% CI)	**P-value**	**Odds ratio (95% CI)**	**P-value**
Use of digoxin (yes vs. no)	4.267 (1.161 – 15.676)	**0.008**	1.617 (1.036 – 3.984)	**0.037**

4.3.3 Survival analysis

Figure 4.1A shows the Kaplan-Meier survival analysis, showing no survival difference between idiopathic DCM patients and familial DCM patients (p=0.739). The mortality rate at the end of a median follow-up period of 5 years was 40% in both groups (Table 4.3). In Figure 4.1B, when the survival of the idiopathic DCM patients is compared to controls matched for age, gender and race in the general South African population, mortality is higher in the idiopathic DCM group (p=0.001).

In Figure 4.1C, the survival of the familial DCM cohort is compared with the matched members of the South African population, and there is a strong trend towards increased mortality in the familial DCM group ($p=0.053$). In Figure 4.1D, the comparison of the transplant-free survival between idiopathic and familial DCM is depicted, showing no difference between the 2 groups ($p=0.986$). In Figure 4.1E, the comparison of the survival free of transplantation between idiopathic DCM patients and controls matched for age, gender and race in the South Africa (SA) population is demonstrated ($p<0.001$); and in Figure 4.1F the comparison of survival free of transplantation in familial DCM patients and controls matched for age, gender and race in the South Africa (SA) population is shown ($p<0.001$). Survival free of transplantation is worse for both groups when compared to the general population.

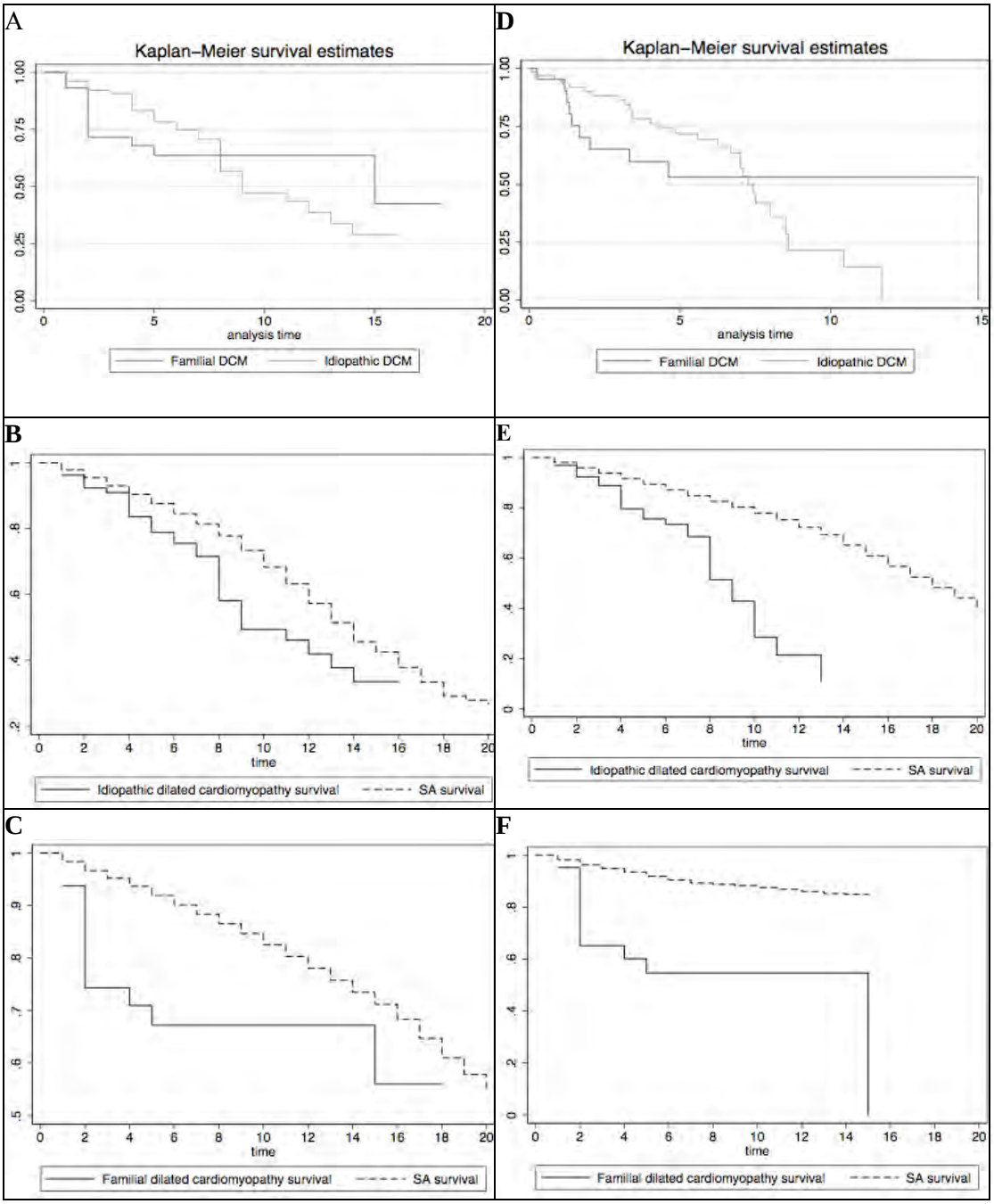

Figure 4.1. Kaplan-Meier survival plots

A, Comparison of survival in idiopathic DCM patients compared to familial DCM patients (p=0.739); B, Comparison of survival in idiopathic DCM patients compared to controls matched for age, gender and race in the South Africa (SA) population (p=0.001); and C, Comparison of survival in familial DCM patients compared to controls matched for age, gender and race in the South Africa (SA) population (p=0.053); D, Comparison of survival free of transplantation in idiopathic DCM patients compared to familial DCM patients (p=0.986); E, Comparison of survival free of transplantation in idiopathic DCM patients compared to controls matched for age, gender and race in the South Africa (SA) population (p<0.001); and F, Comparison of survival free of transplantation in familial DCM patients compared to controls matched for age, gender and race in the South Africa (SA) population (p<0.001).

4.4 Discussion

This study of 80 idiopathic DCM and 40 familial DCM patients has shown a number of important observations. Firstly, despite the more favourable age and LV function of the familial DCM, there was a high mortality of 40% after a median follow-up of five years that was similar to patients with idiopathic DCM. Second, the presence of symptoms of heart failure was the most important clinical predictor of mortality, which was significantly reduced by intervention with OHT. Thirdly, and of concern, the use of digoxin in patients with idiopathic DCM appeared to be associated with excess mortality.

We show that patients with idiopathic DCM have greater LV dimension both in systole and diastole on echocardiography, more radiographic cardiomegaly, as well as lower contractile capacity on echocardiography and cardiac catheterisation. Furthermore, on ECG we found a greater prevalence of pathological Q waves in idiopathic DCM in the absence of coronary artery disease. Similarly, Grünig and colleagues have shown that ECG ST segment and T wave abnormalities occurred more frequently in familial DCM than in idiopathic DCM.[91] However, other authors have failed to demonstrate ECG differences between familial and idiopathic cases of DCM.[75,76,108] ECG features are not sufficiently specific or sensitive to distinguish between familial DCM and idiopathic DCM, and are not useful for clinical classification. In this study, as with most comparisons of familial DCM with idiopathic DCM,[75,80,89] there were no reliable clinical or morphological parameters capable of distinguishing between familial and non-familial forms of DCM, apart from a younger age of onset.[105]

We show that persistent symptoms of heart failure associated with NYHA functional class III or IV features are the most powerful predictor of mortality both in idiopathic and familial DCM.

Similarly, Limongelli and colleagues have shown that, in cohort of 48 adolescents with idiopathic DCM, NYHA functional class III and IV, pro-brain natriuretic peptide and electrocardiographic LA enlargement were the most important predictors of adverse outcomes.[109] Pasotti and co-investigators also reported on a group of 27 consecutive patients with DCM and lamin A/C gene mutations and found that NYHA functional class III and IV and competitive sports were the significant predictors of all cardiovascular events, including death.[110] Other important predictors of mortality in DCM patients, which were not confirmed in this study, include advanced age, protodiastolic gallop, failure of the myopathic ventricle to respond to inotropic stimulation, ventricular arrhythmias, prolonged ECG QRS duration, reduced LV function and late gadolinium enhancement (LGE) on cardiovascular magnetic resonance (CMR) imaging.[111,112]

The annual mortality rate for a patient with heart failure is 10-13%, which is similar to the mortality rate of 50% after a median follow-up period that was found in this study.[113] However, while the rate of progression is variable and influenced by several factors, symptomatic patients experience progressive deterioration, and 10-50% of symptomatic patients with heart failure may die within a year.[114] Hence, it is important that clinicians should pay particular attention to medical management of persistent symptoms in DCM patients, with the aim of escalating management including use of CRT and transplantation, as NYHA class III and IV symptoms remain powerful predictors of mortality. The medical history therefore remains the most important tool for assessment of adequacy of treatment in DCM.

The patients in this study were on good medical treatment, with more than 60% simultaneously receiving an ACE-inhibitor or ARB, a beta-blocker, spironolactone and digoxin. However, we found that beta-blockers and digoxin were more commonly prescribed in the idiopathic DCM group. The increased use of beta-blockers and digoxin in the idiopathic DCM may reflect that this group of patients had more severe disease, as evidenced by greater LVEDD and lower LVEF. It has

been observed by others that patients with familial DCM are less intensively treated with evidence-based therapies for heart failure than patients with sporadic DCM.[89]

Interestingly, digoxin appears to be a significant predictor of mortality in idiopathic DCM, but not in familial DCM patients. Evidence from old clinical trials which were conducted before the introduction of beta-blockers for the treatment of heart failure supports the use of digoxin in patients with LV systolic dysfunction for the treatment of heart failure, particularly in patients with advanced symptoms.[115,116] There is, however, no evidence that digoxin improves survival – and, in fact, may worsen outcomes in DCM patients. The effect of digoxin therapy differs between men and women; digoxin therapy is associated with an increased risk of death from any cause among women with heart failure and depressed LV systolic function.[117] The DIG trial, a study of almost 6800 patients with symptomatic heart failure, LVEF below 45% and who were in sinus rhythm, assigned randomly to receive either digoxin or placebo, showed that after 3 years of follow-up: (1) there was no difference in survival between digoxin and placebo groups; (2) the patients on digoxin had reduced symptoms and hospitalisation for heart failure; (3) patients on digoxin had significant increase in non-heart failure deaths, including deaths from arrhythmia, particularly in women; and (4) that lower serum digoxin levels correlated with survival (the ideal serum level is 0.65-1.0 nmol/l).[118,119] The serum levels of digoxin were not monitored in our study. The results of our analysis suggest that clinicians should refrain from blanket prescription of digoxin for all DCM patients without monitoring of digoxin levels.

The findings of this sub-study have several important implications for clinical practice and research. First, the presence of symptoms of heart failure with mild cardiac structural and functional abnormalities in relatives of patients with DCM is associated with a poor prognosis. Second, the

study emphasizes the need to refer patients with DCM and persistent symptoms of effort intolerance for evaluation for life-saving interventions such as heart transplantation. There is anecdotal evidence that patients with DCM are not referred in a timely manner for OHT in South Africa. Finally, there is a need to review the use of digoxin in patients with heart failure in view of the mortality risk that has been shown in this study. It is prudent to monitor digoxin levels, together with serum potassium and renal function, in all patients in view of the correlation of serum levels with mortality.[119]

While this sub-study has important findings for clinical practice, it also has several limitations. First, generalization of findings from this study may be limited by the relatively small sample size. Second, this study population is dominated by patients with familial DCM that is typical of a tertiary referral clinic.

4.5 Conclusions

In conclusion, DCM due to familial and non-familial causes is associated with a high mortality despite optimal medical and surgical therapy. Patients with persistent symptoms of heart failure despite optimal medical therapy need to be considered and referred for early assessment for OHT and other life-saving interventions.

Chapter 5

Persistent symptoms of heart failure predict mortality in Africans with peripartum cardiomyopathy

5.1 Introduction

There is a paucity of studies of mortality and predictors of outcome in patients with peripartum cardiomyopathy (PPCM) who live in Africa where the condition is endemic.[25,33,120] Sliwa and colleagues have reported a prospective study of 80 patients with PPCM who had a 25% case fatality rate at 2 years of follow-up which was not affected by HIV infection.[120] There are, however, widely varying estimates of the survival rate in patients with PPCM. Amos *et al* observed no mortality in 55 patients followed up over an average of 43 months during a 13 year period,[121] while Brar *et al* described a case fatality rate of 3.3% over a 4.7 year follow-up.[122] Mortality rates of 9.3% were observed by Goland and colleagues in the United States over a mean follow-up period of 19 months,[123] and Duran and co-authors described a mortality of 30% in 33 in Turkish patients followed-up between 1995 and 2007.[124] These varying estimates may be related to differences in case selection and possibly the inclusion of patients with hypertension-related heart failure.[23]

The aims of this study were to establish the case fatality rate and predictors of death in a prospective cohort of consecutive patients with PPCM who were identified over 14 years at a

tertiary hospital. Patients enrolled into the study were evaluated for PPCM at the Cardiac Clinic, GSH.

5.2 Methods

5.2.1 Study design

The sub-study was designed as a hospital-based study of the case fatality rate and predictors of death in a prospective cohort of consecutive patients with PPCM who were identified over 14 years at a tertiary hospital - GSH, Cape Town, South Africa. We reviewed the medical records of all patients diagnosed with idiopathic PPCM, evaluated at GSH Cardiac Clinic from February 1, 1996 and December 31, 2009.

5.2.2 Study population

The patients in this study were seen in a dedicated cardiomyopathy clinic. All patients included in the study were diagnosed using established criteria for PPCM,[22] based on evidenced by clinical heart failure associated with a LV dilatation and a LVEF less than 45% on echocardiography, either in the last trimester of pregnancy or in the first 5 months post-delivery. The selection of the patients for inclusion in this study has been described in detail before.[213]

5.2.3 Data collection

All patients had comprehensive clinical assessment. Information collected at the time of physical examination included history of medical co-morbidity, medications used, history of alcohol and tobacco use. Important clinical parameters including the pulse, blood pressure, oedema, the height of the jugular venous pressure, presence of murmurs and crepitations were recorded. Clinical

assessment was complemented by chest radiography, electrocardiography, and detailed two-dimensional and Doppler colour-flow echocardiography. Normal values for echocardiographic measurement were based on age and body-surface area.

5.2.4 Statistical analysis

Simple descriptive statistics were used for data interpretation and to draw inferences about the population of patients studied. Results of quantitative traits are given as means ± SD. Categorical traits are represented as number and percentage. Pearson's *chi-square* for categorical variables, Student's *t* test for continuous variables or Fisher's exact test for non-parametric distributions were used to compare the relative frequency of characteristics between individuals. All P values were two-sided; and P values above 0.05 were considered not to indicate statistical significance. Univariate and multivariate logistical regression analysis was used to determine predictors of death.

5.2.5 Ethical considerations

The study was approved by the University of Cape Town Faculty of Health Sciences Human Research Ethics Committee (REC Ref No. 197/96). All participants gave written informed consent to participate in the study. All eligible patients were asked for permission to invite their first-degree relatives (parents, siblings and children) to participate in the study.

5.3 Results

5.3.1 Baseline characteristics and pregnancy features

Table 5.1 shows the demographic and clinical characteristics of the 30 participants in this study. The mean age was 31.45 ± 7.48 years. Three (10%) patients were expecting twins in the index pregnancy. All the patients developed symptoms of congestive heart failure in the postpartum

period: 9 (30.0%) in the first week after delivery; 7 (23.3%) between the end of the first week and first month postpartum; and 14 (46.7%) after the first month but before the end of the fifth month in the puerperium. The left ventricular end systolic dimension (LVESD) and left ventricular end diastolic dimension (LVEDD) were 6.85 ± 0.74 cm and 7.39 ± 1.13 cm, respectively. The mean left ventricular ejection (LVEF) fraction on echocardiography was 23.84 ± 8.33 at baseline and 31.28 ± 11.39 at the last-follow up visit.

5.3.2 Follow-up and outcomes

Study subjects were followed-up for a median of 4.33 years (range 0.16 – 13.8 years), during which 5 (16.7%) patients died. 24 (80%) patients, including those who died, remained in chronic heart failure throughout the period of study (Table 2). Other complications included intra-cardiac thrombus formation (16.7%), pulmonary hypertension (13.3%), atrial fibrillation (10.0%), and stroke (6.7%). One patient had cardiac resynchronisation therapy and another patient had orthotopic heart transplantation. At the last visit, 18 (72.0%) patients had NYHA functional class I and II symptoms, and 7 (28.0%) had class III and IV symptoms. 8 (26.7%) women improved their LVEF by more than 10% during the study period, while the rest either remained the same or worsened. However, none of these PPCM patients recovered their LVEF to greater than 50%. On multivariate logistic regression analysis, NYHA functional class III/ IV status at last visit was the only significant independent predictor of mortality (OR 3.11 [C.I. 1.33 − 11.98], p=0.047).

Table 5.1. Clinical characteristics of patients diagnosed with PPCM

Clinical characteristics of PPCM patients (N=30)	
Age (years) ± SD	31.45 ± 7.48
Age > 30 years (%)	17 (56.7)
Unemployment	10 (33.3)
Parity ± SD	2.40 ± 0.73
Gravidity ± SD	2.42 ± 0.68
Multipara (%)	21 (70.0)
Twin pregnancy (%)	3 (10)
Tocolytic therapy (%)	1 (3.3)
Tobacco smoking Never smoker Former smoker Current smoker	 21 (70.0) 2 (6.7) 7 (23.3)
Diagnosis delay in weeks ± SD	1.43 ± 0.54
CTR > 50% (%)	29 (97.6)
Heart rate (bpm) ± SD	86.3 ± 17.82
QRS duration (ms) ± SD	109.50 ± 12.17
LVESD (cm) ± SD	6.85 ± 0.74
LVEDD (cm) ± SD	7.39 ± 1.13
LVEF baseline (%) ± SD	23.84 ± 8.33
LVEF at last follow-up (%) ± SD	31.28 ± 11.39
Heart failure therapy at last follow-up visit (%) Furosemide ACE-I/ARB B-blocker Digoxin Spironolactone Warfarin	 30 (100.0) 30 (100.0) 28 (93.3) 23 (76.7) 27 (90.0) 9 (30.0)

ACE-I, angiotensin con verting enzyme; ARB, angiotensin receptor blocker; LVEDD, left ventricular end-diastolic dimension; LVESD, left ventricular end-systolic dimension; LVEF, left ventricular ejection fraction; PPCM, peripartum cardiomyopathy

Table 5.2. Outcomes in PPCM patients during follow-up of 14 years (median follow-up of 4.3 years)

Outcomes in PPCM patients	***Proportion affected***
Death	5 (16.7)
Chronic heart failure	24 (80.0)
Atrial fibrillation	3 (10)
Cardiac resynchronisation therapy	1 (3.3)
Intra-cardiac thrombus	5 (16.7)
Stroke	2 (6.7)
Pulmonary hypertension	4 (13.3)
Orthotopic heart transplant	1 (3.3)
NYHA functional class at last visit Class 1 and 2 Class 3 and 4	 18 (72) 7 (28)

NYHA, New York Heart Association

5.4 Discussion

To the best of our knowledge, we present the longest follow-up study of PPCM in Africans which provides four insights. First, we show that the case fatality rate is about 16.7% over a median of 4.3 years in patients on modern medical therapy, a finding that is consistent with the largest study of outcome in PPCM.[123] The outcome in this study is better than the series of Sliwa *et al* in comparable African women from Johannesburg where a case fatality rate of 25% was reported at 2 years of follow-up.[120] The reasons for the differences in outcome between the Cape Town and Johannesburg cohorts are not clear. Second, we confirm the association of PPCM with adverse outcomes including chronic progressive congestive heart failure, arrhythmia, thromboembolic complications (affecting the cerebral, coronary, mesenteric, peripheral and pulmonary vascular beds), sudden cardiac death within 3 months from diagnosis, and premature cardiovascular mortality.[23,124] Third, we show in this study that chronic heart failure was common, occurring in 80% of patients studied, and with virtually no normalization in left ventricular dysfunction. By contrast, American women with PPCM show recovery in almost 50% of cases.[125] Our findings

support the recommendation for long-term treatment and monitoring of patients with PPCM.[25,120] Finally, we found the persistence of symptoms of heart failure to be the strongest predictor of death in patients with PPCM. The persistence of symptoms in patients with PPCM on full medical therapy should therefore prompt the consideration of additional interventions, such as early evaluation for orthotopic heart transplantation.

The major limitation of this study is the relatively small sample size of participants. PPCM is a relatively uncommon condition, and the numbers studied are typical of other contemporary studies of this condition.

5.5 Conclusions

PPCM still has a high mortality, particularly in those women with persistent symptoms of heart failure. PPCM is often associated with adverse outcomes including chronic progressive congestive heart failure, arrhythmia, thromboembolic complications (affecting the cerebral, coronary, mesenteric, peripheral and pulmonary vascular beds), sudden cardiac death within three months from diagnosis, and premature cardiovascular mortality. Failure of normalisation of LV function is common. The persistence of symptoms of heart failure is the strongest predictor of death in patients with PPCM. The persistence of symptoms in patients with PPCM on full medical therapy should therefore prompt the consideration of additional interventions, such as early evaluation for OHT.

Chapter 6

Pregnancy-associated heart failure: A comparison of clinical presentation and outcome between hypertensive heart failure of pregnancy and idiopathic peripartum cardiomyopathy

6.1 Introduction

PPCM is defined as a myocardial disorder of unknown cause, characterised by marked impairment of LV systolic function, with development of heart failure towards the end of pregnancy and in the months following delivery, in women without pre-existing heart disease, and in the absence of any other identifiable cause of peripartum heart failure.[21,22] Although the aetiology of PPCM is poorly understood,[23,126] many authorities consider pregnancy-induced hypertension (PIH) to be a risk factor for PPCM.[20,126-129] Furthermore, it has been postulated that hypertensive heart failure of pregnancy (HHFP), defined as the occurrence of peripartum heart failure in association with any form of hypertension, and PPCM may represent a spectrum of the same disease which has a common pathophysiological pathway.[130]

Our group has proposed a clear case definition of PPCM, noting that PPCM should be a diagnosis of exclusion that precludes all other known causes of peripartum heart failure, including hypertension.[23,69] Since the seminal description of PPCM by Demakis in 1971, where preeclampsia

was found in 22% of the 27 patients studied,[20] it has been unclear what the role of PIH in PPCM is. Indeed, many authors have described great variability in the prevalence of PIH in PPCM, with preeclamptic patients accounting from 15 to 89% of PPCM patients reported in the different studies.[20,70,121,123,127-129,131-133] It is has been suggested that the inclusion of patients with varying degrees of gestational hypertension, in the index as well as previous pregnancies, has contributed significantly to the discrepancy in reported characteristics of PPCM.[22,23,134]

We have performed a study of the clinical characteristics and outcome of a consecutive series of patients with a new diagnosis of pregnancy-associated heart failure occurring in the last trimester of pregnancy or puerperium in Cape Town. We investigated whether there were significant differences in time of onset of symptoms, clinical profile, and outcome between HHFP cases compared to patients with unexplained PPCM, in order to assess whether they can be considered to be a spectrum of the same disease, or whether they should be classified differently.

6.2 Methods

6.2.1. Study design

This was a prospective, longitudinal hospital-based case-comparison study of HHFP to PPCM, in patients presenting with heart failure between the last month of pregnancy and the fifth postpartum month, who had been referred to the Cardiac Clinic at GSH in Cape Town, South Africa.

We invited clinicians to refer patients with a new diagnosis of heart failure occurring in the last trimester or puerperium for enrollment into a study of risk factors of PPCM. The exclusion criteria were a known cardiac lesion; in particular, valvular heart disease, previous anthracycline exposure,

ischaemic heart disease, a congenital heart lesion, and metabolic and systemic disorders with cardiovascular sequelae including diabetes mellitus and thyroid disease. Patients with any form of hypertension in pregnancy (including PIH) were included in the study; the latter patients were classified as HHFP. The study participants were between February 1, 1996 and December 31, 2009 in a specialist cardiomyopathy clinic. The median follow-up for PPCM was 3.5 years and 6 years for HHFP.

6.2.2. Definition of HHFP and PPCM

The diagnosis of HHFP was based on the presence of clinical heart failure associated with any form of hypertension (i.e., chronic hypertension, gestational proteinuric hypertension, preeclampsia, eclampsia and postpartum hypertension), occurring in women between the last month of pregnancy and the first 5 months of the postpartum, in the absence of pre-existing heart disease, and any other identifiable cause of peripartum heart failure besides hypertension. Even though most of this group of study subjects had evidence of depressed systolic function (LVEF less than 45%) shortly after delivery, on echocardiography, there was no restriction on subject selection based on echocardiographic parameters.

The standard definition of PPCM was applied; the patients with PPCM needed to fulfill echocardiographic criteria of a left ventricular ejection fraction (LVEF) below 45%, left ventricular fractional shortening below 28%, and a left ventricular end diastolic dimension greater than 5.5cm or greater than 2.7cm/m^2.[21] Patients with any form of hypertension, preeclampsia or eclampsia were excluded from the PPCM group.

6.2.3. Data collection

There were 36 patients with PPCM that were enrolled in the study and 6 of these were subsequently lost to follow-up, and were not included in the analysis. All patients with a diagnosis of HHFP were included in the analysis. All patients had comprehensive clinical assessment, complemented by chest radiography, electrocardiography (ECG), two-dimensional and Doppler colour-flow echocardiography, and cardiac catheterisation, when appropriate. Blood was taken for full blood count, serum creatinine, urea and electrolytes. The primary imaging modality used to confirm the diagnosis was transthoracic two-dimensional and Doppler echocardiography. As the analysis period for the study is over 14 years, the echocardiographic assessments were performed by different cardiologists and sonographers at different time points in the follow-up, and some of these echocardiographic studies were incomplete. In the end, we were able to establish the vital status of 30 PPCM and 53 HHFP patients at the end of the follow-up period.

6.2.4. Statistical analysis

Results of quantitative measurements are given as means ± SD. Categorical traits are represented as number and percentage. Pearson's chi-square or Fisher's exact test were used to compare the relative frequency of characteristics between the two groups of patients. All P values are two-sided; and P values < 0.05 are considered to be statistically significant. Survival analysis was performed using Kaplan-Meier plots.

6.2.5. Ethical considerations

This sub-study was approved by the University of Cape Town Faculty of Health Sciences Human Research Ethics Committee. All patients gave informed consent for participation in the study, and the study complies with the Declaration of Helsinki.

6.3 Results

6.3.1. Baseline characteristics

A total of 83 female patients were included in this study (shown in Table 6.1). PPCM patients had an average age of 31.5 ± 7.5 years and HHFP patients had a similar mean age of 29.6 ± 6.6 years ($p = 0.22$). Nine (30%) of PPCM patients developed heart failure in the first week after delivery, 7 (23.3%) after the first week but before the end of the first postpartum month, and 14 (46.7%) after the first month, but before the end of fifth postpartum month. In contrast, the HHFP patients presented earlier, with 45 (84.9%) developing heart failure in the last month of pregnancy, 6 (11.3%) in the first postpartum week, and 2 (3.8%) between the first and fifth postpartum months ($p<0.001$) (as depicted in Figure 6.1). Twin pregnancy occurred more commonly in PPCM patients ($p = 0.04$), as did smoking ($p = 0.02$). A family history of hypertension ($p<0.001$) and a history of hypertension in a previous pregnancy ($p<0.001$) were found more commonly in HHFP patients.

Figure 6.1. Clinical characteristics of patients with peripartum cardiomyopathy (PPCM) and hypertensive heart failure of pregnancy (HHFP) at the initial presentation with heart failure

Clinical characteristics	PPCM (N=30)	HHFP (N=53*)	P-value
Ethnicity			
Black/African	18 (60.0)	30 (56.6)	0.820
Coloured/Mixed ancestry	12 (40.0)	23 (43.4)	
Age at diagnosis	31.5 ± 7.5	29.6 ± 6.6	0.223
Onset of symptoms in relation to pregnancy			
Last trimester	0 (0)	45 (84.9)	<0.001
Within 1st week after delivery	9 (30.0)	6 (11.3)	
> 1st week,<1st month after delivery	7 (23.3)	0 (0)	
> 1st month,<5th month after delivery	14 (46.7)	2 (3.8)	
Twin pregnancy	3 (10.0)	0 (0)	0.04
Family history of hypertension	3 (10.0)	35 (66.0)	<0.001
History of hypertension in previous pregnancy	0 (0)	17 (32.1)	<0.001
NYHA FC at presentation			
Class 1 and 2	10 (33.3)	9 (16.9)	0.163
Class 3 and 4	20 (66.7)	44 (83.1)	
Pedal oedema	24 (80.0)	19 (35.8)	<0.001
Parity	2.4 ± 0.7	2.2 ± 0.6	0.591
Gravidity	2.4 ± 0.7	2.2 ± 0.6	0.595
Smoking			
Never smoker	21 (70.0)	33 (62.3)	0.024
Former smoker	2 (6.7)	15 (28.3)	
Current smoker	7 (23.3)	5 (9.4)	
Alcohol			
Never drinker	23 (76.7)	45 (89.4)	0.244
Former drinker/moderate use	4 (13.3)	7 (13.2)	
Excessive intake	3 (10.0)	1 (1.9)	
HIV seropositive	3 (10.0)	6 (11.3)	0.582
Delay from symptom onset to clinical assessment (months)	2.7 ± 1.4	1.1 ± 0.3	<0.001
Systolic blood pressure	105.9 ± 16.2	162.3 ± 28.4	0.003
Diastolic blood pressure	63.5 ± 9.6	105.0 ± 12.1	<0.001
Basal rales	16 (53.3)	41 (77.4)	0.007
Murmur			
No murmur	10 (33.3)	35 (66.0)	0.006
MR	13 (43.3)	14 (24.6)	
MR + TR	7 (23.3)	3 (5.7)	
ESM	0 (0)	1 (1.9)	

(*48 [90.6%] of the 53 HHFP patients had a diagnosis of preeclampsia; NYHA FC=New York Heart Association functional class; MR, mitral regurgitation; TR, tricuspid regurgitation; ESM, ejection systolic murmur)

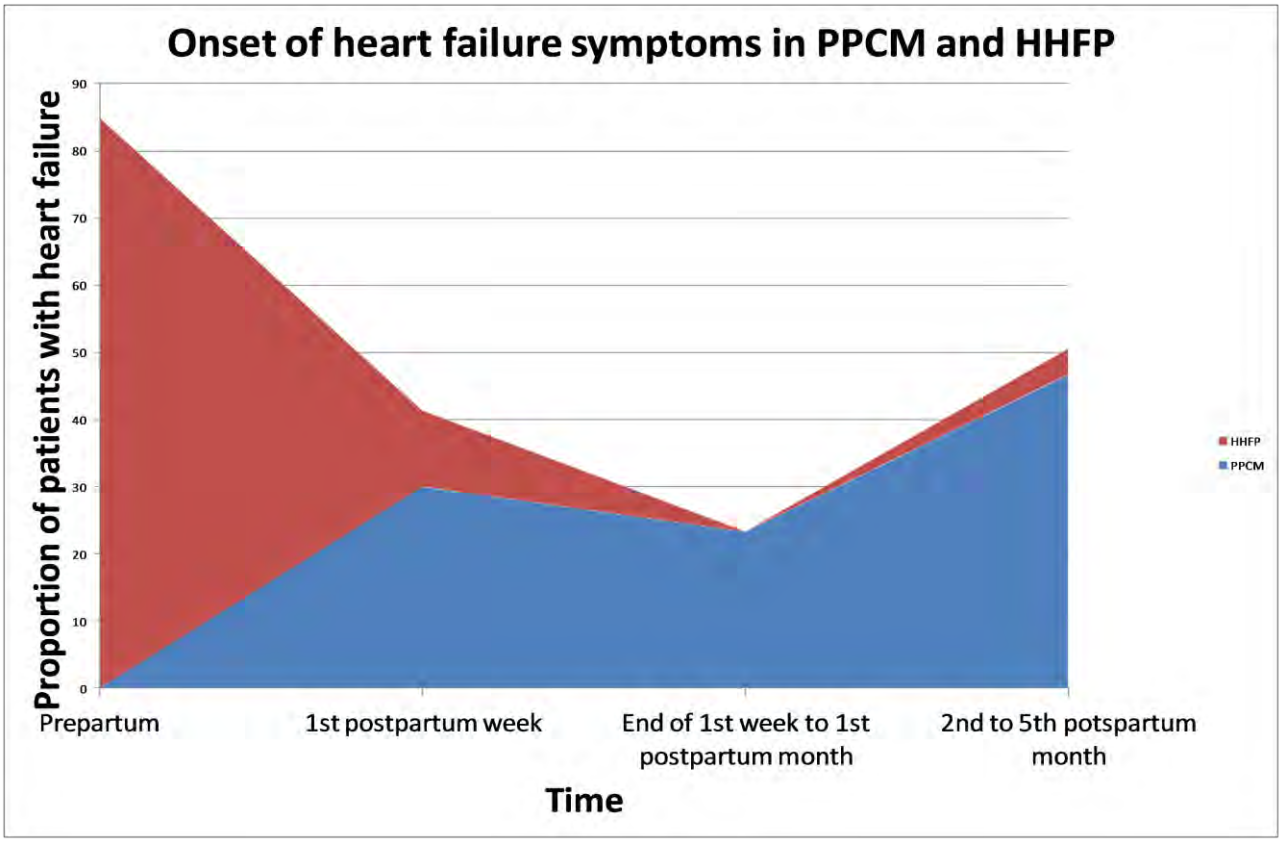

Figure 6.1. Onset of heart failure symptoms in hypertensive heart failure of pregnancy and peripartum cardiomyopathy

6.3.2. Clinical characteristics at presentation

Three (10%) of the PPCM patients and 6 (11.3%) of the HHFP patients were HIV-infected (p = 0.582). A sensitivity analysis revealed that there were no differences between patients with and without HIV infection. The systolic and diastolic blood pressures measurements were 105.9 ± 16.2 mmHg and 63.5 ± 9.6 mmHg in PPCM patients and 162.3 ± 28.4 mmHg and 105.0 ± 12.1 mmHg in HHFP patients (p = 0.003 and p<0.001, respectively). Basal rales were detected in 16 (53.3%) of PPCM patients and 41 (77.4%) of HHFP patients (p = 0.007). Peripheral oedema was present in 80% of PPCM patients compared to 35.8% of HHFP patients (p<0.001).

6.3.3. Radiographic, electrocardiographic and echocardiographic features

A cardiothoracic ratio (CTR) greater than 50% was found more commonly in PPCM compared to HHFP patients ($p<0.001$), as shown in Table 6.2. Radiographic evidence of pulmonary oedema was noted more frequently in HHFP patients, in keeping with the clinical detection of rales ($p = 0.010$). Higher heart rate was found in HHFP patients ($p = 0.014$). On electrocardiography (ECG), atrial fibrillation ($p = 0.028$), QRS abnormalities ($p = 0.001$), relatively longer QRS duration ($p<0.001$), left atrial hypertrophy ($p = 0.030$), and left bundle brunch block ($p = 0.002$), and T wave inversion ($p<0.001$) were detected more commonly in PPCM patients than in HHFP patients. As expected, left ventricular hypertrophy (LVH) ($p = 0.021$) was seen more frequently in HHFP patients. On echocardiography, patients with HHFP had greater ventricular septal thickness in diastole ($p = 0.013$) as well as greater LV posterior free wall thickness in systole ($p = 0.002$). PPCM patients had larger LV dimensions both in systole and diastole ($p<0.001$ and $p<0.001$, respectively), as well as lower LVEF and LV fractional shortening ($p<0.001$ and $p<0.001$, respectively) compared to HHFP patients.

6.3.4 Medical management at follow-up

With regards to heart failure therapy, significantly more PPCM patients were receiving furosemide ($p<0.001$), angiotensin converting enzyme-inhibitor (ACE-I)/angiotensin receptor blocker (ARB) ($p<0.001$), β-blockers ($p<0.001$), spironolactone ($p<0.001$) and digoxin ($p<0.001$) compared to HHFP patients (Figure 6.2). However, more HHFP patients were on calcium channel blockers (CCB) for treatment of hypertension than PPCM ($p = 0.014$). Warfarin was prescribed more commonly for consequent atrial fibrillation, and LV thrombus for PPCM patients ($p = 0.030$).

Table 6.2. Radiologic, electrocardiographic, and echocardiographic findings at initial presentation with heart failure

Radiologic, electrocardiographic, echocardiographic and laboratory findings	PPCM (N=30)	HHFP (N=53)	P-value
Radiographic cardiothoracic ratio>50%	29 (96.7)	22 (41.5)	<0.001
Radiographic pulmonary oedema	16 (53.3)	45 (89.4)	0.010
ECG heart rate	86.3±17.8	106.9±27.4	0.014
ECG QRS abnormalities	8 (27.6)	1 (1.9)	0.001
ECG voltage abnormality	8 (27.6)	31 (58.5)	0.816
ECG left anterior hemiblock	9 (30.0)	5 (9.4)	0.03
ECG Q wave	5 (16.7)	5 (9.4)	0.484
ECG left bundle branch block	6 (20.0)	0 (0)	0.002
ECG LVH	8 (26.7)	29 (54.7)	0.021
ECG T wave inversion	22 (73.3)	13 (24.5)	<0.001
Atrial fibrillation	3 (10.0)	0 (0)	0.028
QRS duration	109.5±12.17	89.8±10.26	<0.001
Echo interventricular septum (diastole)	0.9 ± 0.2	1.2 ± 0.2	0.013
Echo LV posterior wall (systole)	1.2 ± 0.3	1.4 ± 0.3	0.002
Echo LV end-systolic diameter (diastole)	6.8 ± 0.7	3.5 ± 0.6	<0.001
Echo LV end-diastolic dimension (diastole)	7.4 ± 1.1	5.1 ± 0.9	<0.001
Echo LV ejection fraction	23.8 ± 8.3	49.9 ± 18.7	<0.001
Echo LV fractional shortening	11.6 ± 4.1	26.2 ± 3.2	<0.001

ECG, electrocardiographic; Echo, Echocardiographic; LV, left ventricle, left ventricular; LVH, left ventricular hypertrophy

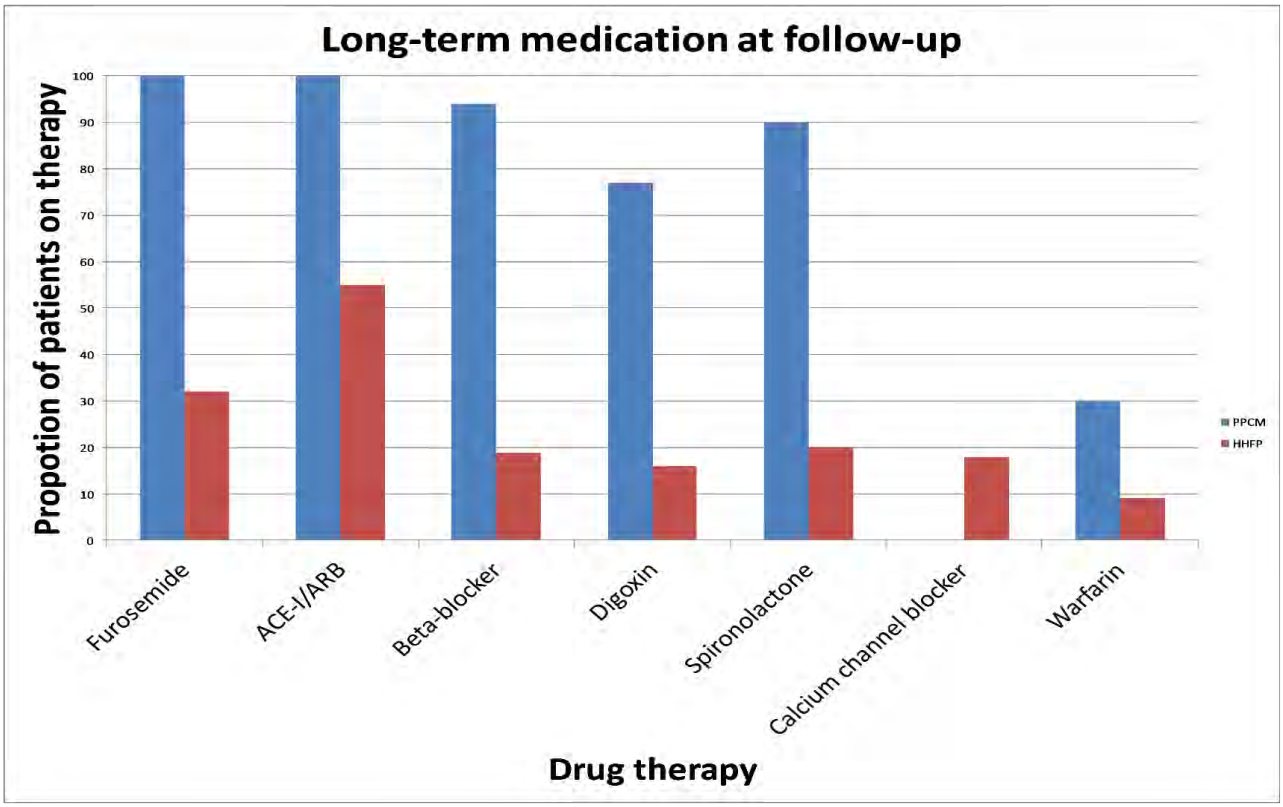

Figure 6.2. Medication prescribed at last follow-up visit

6.3.5. Survival analysis

There were 5 deaths in PPCM patients during the 14 years of follow-up, while there were no deaths in the HHFP group ($p = 0.005$). A Kaplan-Meier analysis of survival stratified according to PPCM versus HHFP at last follow-up visit ($p<0.001$) is shown in Figure 6.3. Table 6.3 shows that chronic heart failure was more common in PPCM patients at the last follow-up visit ($p<0.001$). Similarly, intra-cardiac thrombus ($p = 0.014$) and development of pulmonary hypertension ($p = 0.022$) were commoner in PPCM patients. At the most recent hospital visit, 18 (72.0%) of PPCM patients were in NYHA functional class I and II and 7 (28.0%) were in functional class III and IV compared to 52 (98.1%) of HHFP patients with class I and II symptoms and 1 (1.9%) with class III and IV symptoms ($p<0.001$).

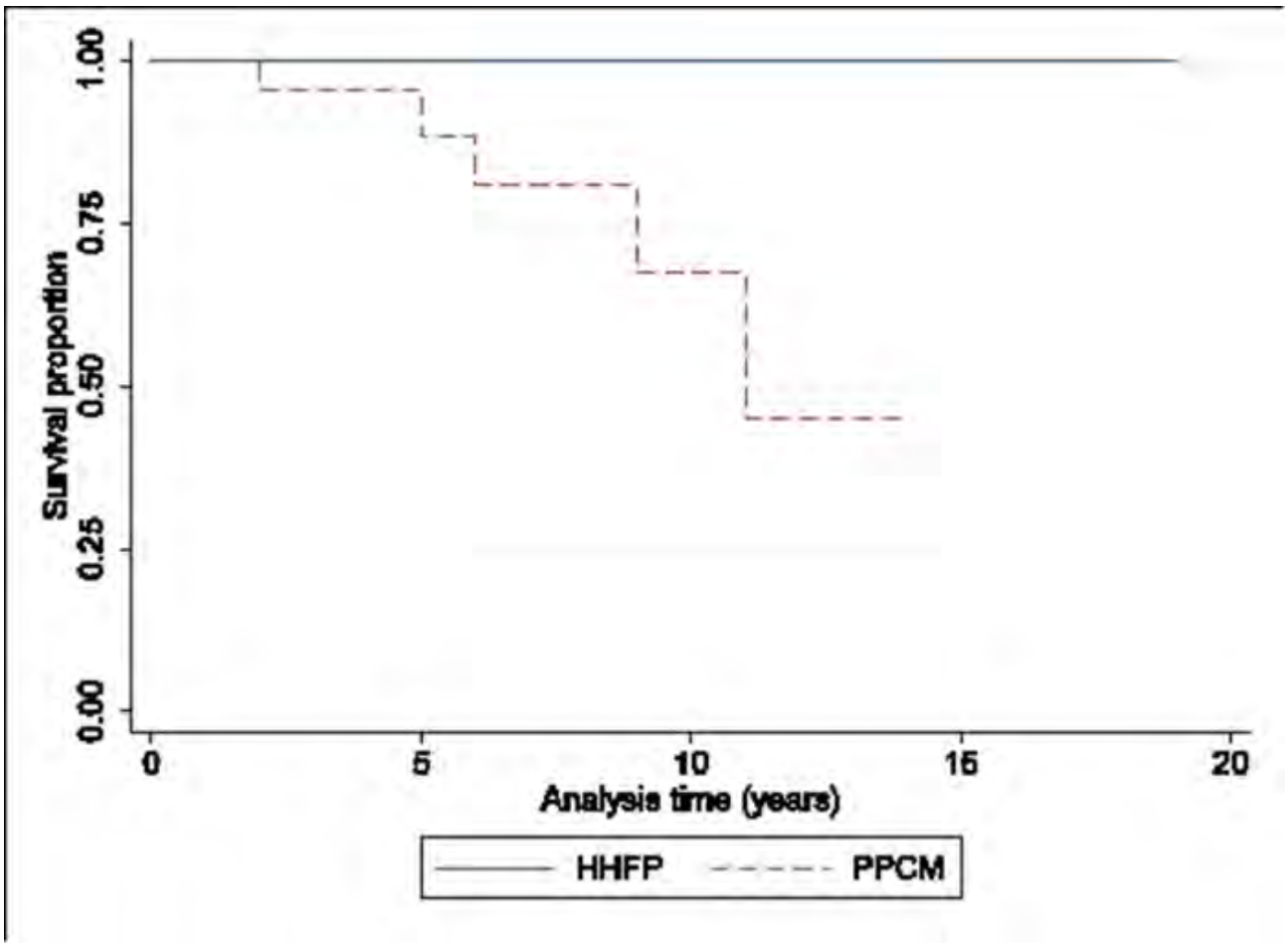

Figure 6.3 Kaplan-Meier survival plot for hypertensive heart failure of pregnancy and peripartum cardiomyopathy patients over 14 years

Table 6.3. Survival and status at last follow-up for the PPCM and HHFP patients studied

	PPCM (N=30)	**HHFP (N=53)**	**P-value**
Median duration of follow-up	3.5 years	6 years	0.02
Death	5 (16.7)	0 (0)	0.005
Chronic heart failure	24 (80.0)	8 (15.1)	<0.001
Cardiac resynchronization therapy	1 (3.3)	0 (0)	0.361
Intracardiac thrombus	5 (16.7)	0 (0)	0.014
Stroke	2 (6.7)	0 (0)	0.128
Pulmonary hypertension	4 (13.3)	1 (1.9)	0.022
Heart transplantation	1 (3.3)	0 (0)	0.361
NYHA FC at last visit Class 1 and 2 Class 3 and 4	 18 (72.0) 7 (28.0)	 52 (98.1) 1 (1.9)	 <0.001

NYHA FC, New York Heart Association Functional Class

6.4 Discussion

We present the results of a case comparison study of the clinical features and outcome of pregnancy-associated heart failure with or without hypertension in patients with no history of structural heart disease and show that in our centre unexplained peripartum cardiomyopathy is exclusively a postpartum disease that is associated with distinct clinical features of heart muscle disease (such as left bundle branch block and atrial fibrillation), chronic heart failure, and a fatal outcome in a proportion of cases. By contrast, HHFP is predominantly an antepartum disease that presents mainly with pulmonary oedema, left ventricular hypertrophy (LVH), and a reversible form of heart failure in the overwhelming majority of cases.

We have found that PPCM to be a postpartum condition, with 53% of PPCM patients presenting within the first month of the puerperium and the other 47% presenting within the subsequent four postpartum months, in contrast to HHFP, where 85% of patients presented with heart failure in the last month before delivery. Our findings are similar to those of another South African study which found that heart failure symptoms develop in the postpartum period in 100% of PPCM patients.[25] Summarising pooled data from 419 cases of PPCM, Lampert and Lang, demonstrated that 78% of PPCM cases developed symptoms in the first four months postpartum, while 9% had their onset of symptoms in the last antepartum month, and 13% either developed symptoms before one month antepartum or after four months postpartum.[135] In the main, however, studies comprising a greater proportion of patients with preeclampsia have documented greater frequency of onset of PPCM in the last month of pregnancy.[120,124] By comparison, studies with fewer cases of pregnancy-associated hypertension have reported a largely postpartum onset of PPCM.[22,20,129,136]

A family history of hypertension was found more commonly in HHFP patients compared to PPCM patients; and in this study, a history of previous hypertension in pregnancy was present in a third of patients with HHFP and absent in those with PPCM. Genetic factors play a role in the development of pregnancy-induced hypertension (PIH), with both maternal and paternal genetic contributions being important.[137,138] Genetic predisposition to PIH/preeclampsia is suggested by the observation that primigravid women with a family history of preeclampsia (i.e., affected mother or sister) have a 2- to 5-fold increased risk of developing the disease than primigravid women without such a history.[139] Furthermore, the spouse of men who are the product of a pregnancy complicated by preeclampsia are more likely to develop preeclampsia than spouses of men without such a history.[140]

Features of congestive heart failure (pedal oedema, elevated jugular venous pressure, and basal rales) were found in up to 80% of PPCM patients, but only in 35.8% of HHFP patients at presentation. In contrast, isolated pulmonary oedema was one of the main clinical and radiologic findings in patients with HHFP in this study. The association of PIH/preeclampsia with isolated pulmonary oedema is well-established.[141,142] Differences in clinical presentation between PPCM and HHFP may also be partly explained on the basis of volume overload in HHFP patients with systolic and diastolic dysfunction.[143] The pathophysiology of pulmonary oedema in preeclampsia is unclear, but is presumed to be due to a combination of microangiopathy, capillary leak and sometimes iatrogenic fluid overload.[144] PPCM, on the other hand, results from development of primary myocardial dysfunction leading to congestive heart failure,[23,126] and has been shown recently, in a mouse model, to be associated with defective cathepsin-D mediated cleavage of prolactin into a 16-kDa form, which is both pro-apoptotic and anti-angiogenic.[145] PPCM may also have genetic underpinnings, and recent reports have supported the contention that PPCM may be part familial dilated cardiomyopathy.[66,105]

Multiple differences were noted on ECG in the comparison between PPCM and HHFP. First, LVH was found to occur more commonly in HHFP than in PPCM. PIH represents a model of acute pressure overload that may induce dramatic changes in LV structure and function.[146] Other important ECG findings from our study included the observation that QRS abnormalities, left atrial hypertrophy, left bundle branch block (LBBB), atrial fibrillation, non-specific T wave inversion and longer QRS duration occurred more frequently in patients with PPCM compared to those with HHFP. Similar findings have been reported previously: repolarisation changes have been reported in 47.3%[147] and LBBB to occur in 10% of PPCM patients.[148] Davidson and Parry found arrhythmias in 2%, and LBBB in 5% of patients with peripartum heart failure, while LVH was present in 26% and T wave changes found in 15%.[149] In a study of 97 PPCM patients from South Africa, LVH was demonstrated in 66% and ST segment and T wave abnormalities noted in 96%.[29] These ECG changes are associated with increased cardiovascular morbidity and mortality in patients with impaired LV function, and QRS duration has been shown to be strongly associated with atrial fibrillation and adverse outcome in patients with cardiomyopathy.[150] The electrocardiographic findings in our study may signify greater myocardial injury in patients with PPCM compared to those with HHFP. While the ECG of most women with PPCM is usually abnormal,[22,151-154] there are no ECG changes that are sufficiently sensitive or specific for the diagnosis of PPCM,[124] nor are there ECG characteristics that serve as a differentiator between PPCM and HHFP.

A higher heart rate and blood pressure were found in patients with HHFP compared to those with PPCM. The finding of increased and/or highly variable pulse rates and elevated arterial pressures in PIH/preeclampsia has been described, and is thought to partly reflect disturbed neural control of heart rate and blood pressure, as a result of impaired sympathetic and parasympathetic nervous system activity in PIH.[155] Maladaptation of the maternal cardiovascular system in PIH/preeclampsia

is manifested as a lack of physiological decline in cardiovascular oscillations of heart rate and blood pressure.[156]

On echocardiography, we found the interventricular septum (IVS) thickness and LV posterior free wall (LVPW) thickness to be increased in HHFP compared to PPCM, likely reflecting LV adaptation to increased wall stress from elevated blood pressure. Similarly, IVS and LVPW thickness were increased in PIH/preeclampsia patients compared with normal controls.[157] PIH was associated with an abnormal LV geometric pattern in 62% of 106 patients studied, of which 42% had eccentric hypertrophy, 17% had concentric remodeling, and 5% had concentric hypertrophy.[158] Again, likely reflecting greater myocardial injury, the LVEF and LV fractional shortening were lower in PPCM patients and were associated with increased LV dimensions in systole and diastole. Similarly, demonstrating impaired LV function (i.e., depressed LVEF, LV cardiac output and stroke volume) and increased LV dimensions have been reported in PPCM by many authors.[22,127-9] Furthermore, the LVEF and LVEDD have been correlated with outcome in PPCM,[123,159] but not in this study.

Finally, we showed that morbidity and mortality was higher in PPCM, with lack of full recovery of cardiac function (average LVEF 23.8% at diagnosis and 31.3% at last follow-up), compared to HHFP patients (average LVEF 49.9% at diagnosis and 68.2% at last follow-up). Similar findings have recently been made by Kamiya and colleagues who examined the clinical profile of Japanese PPCM patients with and without gestational hypertension and found that patients with pregnancy-associated hypertension diagnosed with PPCM had a shorter hospitalisation and higher LVEF at last follow-up when compared to the PPCM patients without hypertension.[159] The mortality for PPCM of 17% over a median of 3.5 years of follow-up is similar to that seen in other countries such as Haiti and Turkey, but much higher than the United States.[134] Chronic heart failure, intra-cardiac thrombus, thromboembolic phenomena and pulmonary hypertension occurred with greater

frequency in patients diagnosed with PPCM. These grave sequelae have been previously reported on in PPCM, and shown to be associated with increased mortality.[123,126,128,160] However, the seriousness of HHFP should not be under-estimated: a recent report on maternal deaths from South Africa showed that preeclampsia/eclampsia and proteinuric hypertension accounted for 83.1% of all PIH-related deaths, and that HHFP was the cause of death in 22.8% of PIH-related deaths in the same period.[161]

The data from this sub-study show that there are significant differences in the time of onset of heart failure, clinical characteristics, and outcomes of patients with HHFP compared to those with truly unexplained PPCM. Hence, these data support the proposal that a history or presence of hypertension in pregnancy should exclude the diagnosis of PPCM, as the two appear to be distinct clinical conditions.[22,23,69] However, this study has a number of limitations including small sample size, being from a single centre, differences in follow-up duration between HHFP and PPCM patients, lack of repeat echocardiography in the majority of HHFP patients at follow-up and the lack of information about repeat pregnancies and the potential for confusion between the symptoms of heart failure and the often similar symptoms of normal pregnancy. It is also important to highlight that, by its very nature, a specialist clinic in a tertiary referral center has a selection bias for the cases that develop chronic PPCM, and may miss cases that demise acutely. Finally, we acknowledge that using strict cut-offs for case definition of PPCM may have led to an underestimate of its prevalence in our setting.

Despite these limitations, this work may contribute to the clarification of the case definition of PPCM by excluding patients with any form of hypertension in this group. We show that HHFP is predominantly an antepartum disease that presents mainly with pulmonary oedema, LVH, and a

reversible form of heart failure in the overwhelming majority of cases. By contrast, at our centre, unexplained PPCM is exclusively a postpartum disease that is associated with distinct clinical features of heart muscle disease (such as LBBB and atrial fibrillation), chronic heart failure, and a fatal outcome in a proportion of cases. Large prospective multicenter studies of peripartum heart failure with and without hypertension are required to confirm the findings of this study.

6.5 Conclusions

In this case comparison study of the clinical features and outcome of pregnancy-associated heart failure with or without hypertension in patients with no history of structural heart disease we found that unexplained peripartum cardiomyopathy is exclusively a postpartum disease that is associated with distinct clinical features of heart muscle disease (such as LBBB and atrial fibrillation), chronic heart failure, and a fatal outcome in some cases. By contrast, HHFP is predominantly an antepartum disease that presents mainly with pulmonary oedema, LVH, and a reversible form of heart failure in the overwhelming majority of cases.

Chapter 7

Clinical features, spectrum of causal genetic mutations, and outcome of hypertrophic cardiomyopathy in South Africans

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7.1 Introduction

Hypertrophic cardiomyopathy (HCM) is defined by the presence of myocardial hypertrophy in the absence of haemodynamic stresses sufficient to account for the degree of hypertrophy (e.g., arterial hypertension and aortic stenosis) and without other secondary causes of cardiac hypertrophy such as amyloidosis and glycogen storage disease.[8] HCM was historically thought to be rare amongst Africans.[32] This impression was reinforced by a study which found HCM to occur in 0.2% of 6680 unselected echocardiograms performed in Tanzania.[31] However, recent echocardiographic studies from the continent have dispelled that myth.[34] For example, in Ghana, HCM has been reported to be the third commonest cardiomyopathy after DCM and endomyocardial fibrosis (EMF).[35] Similarly, in Ethiopia, HCM accounts for 34% of all cardiomyopathies diagnosed on echocardiography.[36] However, there is a dearth of information on the clinical features, genetics and outcome of HCM from the African continent, with a few publications reporting on HCM-causing mutations in South Africans of North-European descent and mixed ancestry.[162-165] To date, there are no data on the genetics of HCM in black Africans.

HCM is a diverse disease with variable phenotypic expression; a substantial proportion of patients live a normal life with minimal risk of sudden cardiac death.[166] However, some patients with or without symptoms may die suddenly even without clinical features of severe LVH.[164] The pattern of LVH in HCM is variable and associated with differences in morbidity and mortality. For instance, apical HCM in the Japanese and North American populations is associated with a benign outcome.[167] The clinical pattern and outcome of HCM in Africans is not known. The aim of this sub-study was to delineate the clinical features, spectrum of disease causing mutations, and outcome of HCM in African patients.

7.2 Methods

7.2.1. Study design

Consecutive patients diagnosed with HCM at the Cardiac Clinic, GSH, Cape Town, South Africa were prospectively enrolled into a longitudinal cohort study of familial cardiomyopathy, from February 1, 1996 to August 31, 2012. The diagnosis of HCM was based on the presence of a hypertrophied, non-dilated LV in the absence of other diseases capable of producing the degree of observed LVH (i.e., LV wall thickness >15mm on echocardiography).[168] Clinical data were collected at 6 monthly visits during the study period.

7.2.2. Measurements and data collection

All patients had comprehensive clinical assessment, complemented by chest radiography, electrocardiography, detailed two-dimensional and Doppler colour-flow echocardiography and cardiac catheterisation, when appropriate. The primary imaging modality used for diagnosis in all

patients was transthoracic two-dimensional and Doppler echocardiography. Patients found to have outflow tract gradients below 40 mmHg underwent Valsalva maneuver. Patients with cardiovascular risk factors, angina or over 40 years old frequently underwent coronary angiography, at the discretion of the attending clinician. A comprehensive database that incorporated patient demographic details, medical history, co-morbidity, medical therapy, clinical, electrographic and echocardiographic details was utilised. Normal values for echocardiographic measurement were based on age and body-surface area as described by Lauer et al.[169]

7.2.3. Genotyping

Peripheral blood was collected from HCM probands for DNA extraction using standard methods. Mutation screening was undertaken by pyrosequencing of the coding regions and exon/intron boundaries of the following 15 genes that are associated with HCM: cardiac myosin-binding protein C (MYBPC3), cardiac β-myosin heavy chain (MYH7), cardiac troponin T type 2 (TNNT2), cardiac troponin I type 3 (TNNI3), regulatory light chain of myosin (MYL2), essential light chain of myosin (MYL3), tropomyosin 1 (TPM1), phospholamban (PLN), α-actin (ACTC1), cysteine and glycine-rich protein 3 (CSRP3), AMP-activated protein kinase (PRKAG2), α-galactosidase (GLA), four-and-a-half LIM domains 1 (FHL1), lamin A/C (LMNA) and lysosome-associated membrane protein 2 (LAMP2) (Table S1).[53]

Exons and intron/exon boundaries (± 10 base pairs) of the 15 cardiomyopathy related genes were amplified by microdroplet polymerase chain reaction (PCR) using RDT 1000 technology (Rain Dance Technologies, Billerica, MA 01821, United States). Libraries were prepared using the Rapid Library 454 FLX protocol, which included adding molecular identifiers to each sample. Samples were pooled and then sequenced using the Roche 454 FLX next-generation sequencing platform.

Samples were processed and analysed using NextGENe version 2.2.0 (SoftGenetics). Prior to analysis, reads were trimmed and low quality reads removed. Reads were aligned to .gbk files and variants seen in <20% annotated. Variants were filtered taking into account coverage, read balance, allele balance and homopolymers. Samples with coverage below 10 were considered failures. Unclassified variants were Sanger sequenced to confirm their presence; known polymorphisms were not Sanger sequenced.

The Cape Town population controls were used to determine the population frequencies of all novel variants identified in the 15 genes. One hundred and ninety five (n=195) anonymous blood donors from the Western Province Blood Transfusion Service (WPBTS) provided consent for blood samples to be taken for DNA extraction. The control DNA consisted of samples from 95 persons of mixed ancestry, 50 black Africans and 50 white South Africans.

7.2.4. Ethical considerations

The study was designed in keeping with the principles of the Helsinki Declaration, and was approved by the University of Cape Town Human Research Ethics Committee. All participants gave informed, written consent to participate in the study.

7.2.5. Statistical analysis

Simple descriptive statistics were used for data interpretation and to draw inferences about the population of patients studied. Results of continuous variables are given as means ± SD. Categorical variables are represented as number and percentage. Pearson's chi-square or Fisher's exact test was used to compare the relative frequency of characteristics between individuals. All P values were two-sided; and P values ≥ 0.05 were considered not to indicate statistical significance. Survival analysis testing between groups was compared using log-rank testing, and the Kaplan-Meier survival curves were constructed using the product-limit method. Age-, gender- and race-adjusted survival curves for the general South African population were derived and compared with the Kaplan-Meier survival rates for the patients with HCM. Analysis included univariate and multivariate regression analysis, with a focus on mortality rather than time to death, thus justifying the use of Cox's proportional hazards model rather logistic regression analysis.

7.3 Results

7.3.1. Clinical characteristics

The study cohort comprised 43 patients with HCM. The clinical characteristics and co-morbid status of the study population at the initial evaluation are shown in Table 7.1 and compared to cohorts from North America, Taiwan and Saudi Arabia. The mean age of HCM patients studied was 38.5 ± 14.3 years; 25 (58.1%) were male. Thirteen (30.2%) were black Africans, and the majority (62.8%) were of mixed ancestry. Twenty six (60.5%) had first degree relatives with HCM and 5 (11.6%) had a family history of sudden cardiac death (SCD) in a first degree relative. Symptoms of palpitations (79.1%), angina (65.1%), fatigue (58.1%), and effort-related breathlessness (55.8%) were frequently reported by the patients. Ten (23.3%) had a New York Heart Association (NYHA) functional capacity of class III at the initial assessment. An ejection systolic murmur was reported in 18 (41.9%) of patients.

Table 7.1. Demographic, clinical, electrocardiographic and echocardiographic features at presentation in patients with hypertrophic cardiomyopathy compared to three large international, contemporary reports from North America, Taiwan and Saudi Arabia

Characteristic	South Africa (n=43)	North America (n=277)	Taiwan (n=163)	Saudi Arabia (n=69)
Medical history				
Age at diagnosis, years	38.5 ± 14.3	47 ± 22	60.9 ± 12.1	42 ± 16
Males	25 (58)	152 (55)	84 (52)	43 (71)
Ethnicity (%)				
Black/African	13 (30)			
White/Caucasian	2 (5)	277 (100)		
Coloured/Mixed ancestry	27 (63)			
Indian ancestry	1 (2)			
Taiwanese			163 (100)	
Arab				69 (100)
First degree relative with HCM	26 (61)	21 (8)	–	2 (5)
Second degree relative with HCM	7 (16)	–	–	–
Has family history of SCD	5 (12)	–	–	4 (9)
NYHA functional class				
Class 1 and 2	33 (77)	–	–	–
Class 3 and 4	10 (23)			
Symptoms		174 (63)		
Fatigue	25 (58)		–	–
Dyspnoea	24 (56)		121 (74)	31 (65)
Palpitations	34 (79)		28 (17)	5 (7)
Angina	28 (65)		111 (68)	–
Presyncope/Syncope	12 (28)		20 (12)	2 (4)
Smoking	19 (31)	–	–	–
Hypertension	12 (28)	–	28 (17)	–
Diabetes	0 (0)	–	29 (18)	–
Alcohol consumption	9 (21)	–	–	–
Dyslipidaemia	6 (14)	–	–	–
Coronary artery disease	3 (7)	–	29 (18)	–
COPD	2 (5)	–	–	–
HIV infection	2 (5)	–	–	–
Medical examination				
Heart rate	71.3 ± 12.7	–	–	–
BP_{sys}	125.8 ± 19.2	–	–	–
BP_{dia}	75.8 ± 11.3	–	–	–
Pedal oedema	5 (11.6)	–	–	–
Ejection systolic murmur	18 (41.9)	–	–	–
Electrocardiographic findings				
Sinus rhythm	39 (90.7)	–	–	–
Atrial fibrillation	4 (9.3)	–	34 (21)	–
QRS abnormalities present	12 (28)	–	–	–

Voltage criteria for LVH	22 (51)	–	137 (84)	60 (87)
Presence of pathological Q waves	12 (28)	–	–	–
T wave inversion	34 (79)	–	108 (66)	–
Left atrial hypertrophy	10 (23)	–	–	–
LBBB	4 (9)	–	–	–
RBBB	2 (5)	–	–	–
PR prolongation	2 (5)	–	–	–
Echocardiographic findings				
LVEDD (cm)	4.1 ± 0.8	–	4.5 ± 0.5	–
LVESD (cm)	2.7 ± 0.6	–	2.4 ± 0.4	–
IVS_{dia} (cm)	1.9 ± 0.7	2.2*	1.9 ± 0.4	–2.1 ± 0.7
IVS_{sys} (cm)	2.1 ± 0.7	–	–	–
$LVPFW_{dia}$ (cm)	1.2 ± 0.4	–	1.1 ± 0.3	1.3 ± 0.4
LVEF (%)	71.5 ± 8.3	–	–	68 ± 13
Left atrial size (cm)	3.5 ± 0.8	–	3.8 ± 0.7	–
SAM	9 (21)	–	80 (49)	39 (57)
LVOT obstruction	12 (28)	–	78 (48)	28 (41)
E/A ratio	1.2 ± 0.4	–	–	1.5 (0.9-2.1)
Pattern of hypertrophy				
Sigmoid	13 (30)	75 (27)	–	8 (12)
Catenoid	23 (53)	92 (33)		29 (42)
Neutral	6 (14)	65 (23)		25 (36)
Apical	1 (2)	5 (2)		7 (10)

All results are means ± standard deviation, unless otherwise indicated.
*No standard deviation given.
BP_{dia}, diastolic blood pressure; BP_{sys}, systolic blood pressure; COPD, chronic obstructive pulmonary disease; E/A, ratio of early (E) to late (A) ventricular filling velocities on Doppler echocardiography; IVS_{dia}, interventricular septal thickness in diastole; IVS_{sys}, interventricular septal thickness in systole; HCM, hypertrophic cardiomyopathy; HIV, human immunodeficiency virus; LBBB, left bundle brunch block morphology on electrocardiography; LVEDD, left ventricular end-diastolic dimension; LVEF, left ventricular ejection fraction; LVESD, left ventricular end-systolic dimension; LVH, left ventricular hypertrophy; $LVPFW_{dia}$, left ventricular posterior free wall thickness in diastole; LVOT, left ventricular outflow tract; NYHA, New York Heart Association functional classification for severity of breathlessness; RBBB, right bundle brunch block morphology on electrocardiography; SAM, systolic anterior motion of the anterior mitral valve leaflet; SCD, sudden cardiac death.

7.3.2 Electrocardiographic and echocardiographic findings

The electrocardiographic and echocardiographic characteristics of the study population are shown in Table 7.1. Four (9.3%) of the HCM patients had atrial fibrillation at diagnosis. On echocardiography, the mean LV septal thickness in diastole, LV ejection fraction (LVEF) and left atrial diameter were 1.9 ± 0.7cm, 71.5 ± 8.3% and 3.5 ± 0.8cm, respectively. LV outflow tract (LVOT) obstruction, with a resting gradient of greater than 10 mmHg, was found in 12

(27.9%) patients. Evidence of diastolic dysfunction was present in the majority of patients, and the mean E/A ratio was 1.2 ± 0.4.

7.3.3. Spectrum of mutations that cause HCM in South Africans

Of the 43 patients diagnosed with HCM, 42 were screened for the common founder mutations previously described in the South African population, and all 42 were found to be negative for these variants.[165] Further molecular genotypic analysis was undertaken in 35 of these HCM patients for 15 cardiomyopathy-associated genes. Of these 35 probands, mutation screening yielded disease-causing mutations in 10 unrelated individuals (28.6%) (Table 7.2). The disease-causing mutations were found in two out of the 15 genes screened, with the majority in MYH7 (n = 6 probands; 60%) and the rest in MYBPC3 (n = 4 probands; 40%). Two of the MYH7 mutations were novel and disease-causing mutations were found in all ethnic groups tested. No single disease-causing mutation occurred in more than one study subject.

There were three genetic variants of unknown significance that were found in MYBPC3 which were not observed in 195 population controls: c.1224-19G>A, c.1790+5G>A, and c.133G>A. In addition, a large number of known polymorphisms were found in 16 probands in MYBPC3 (tmp_esp_11_47355301, rs113941605 and rs113658284), MYH7 (rs149439730, rs45523835, rs145738465, rs202205780, rs61737803, rs146858930, rs36211714, rs45501694, and rs111626355), TNNT2 (rs113471285, and rs115805892), TPM1 (tmp_esp_15_63356347), MYL3 (rs199474709), CSRP3 (rs112848043), FHL1 (rs182106777), PRKAG2 (rs116605521, and rs113234987), GLA (rs151195362), and LMNA (rs12117552, and rs117939448). Two novel single nucleotide polymorphisms were found in MYH7 (c.1368C>T) and PRKAG2 (c.828C>A) in two different probands.

Table 7.2. Disease causing mutations found in 10 unrelated index cases with hypertrophic cardiomyopathy

Index case ID	Ethnicity	Reported previously ?	Gene	Exon	Nucleotide and Amino acid change	Type of mutation	Reference
HCM1.1	Indian	Yes	MYH7	5	c.611G>A (p.R204H)	Missense	Richard et al 2003[18]
HCM4.1	Mixed ancestry	No	MYH7	20	c.2282C>A (p.T761N)	Missense	Novel
HCM7.1	Mixed ancestry	Yes	MYH7	31	c.4258C>T (p.R1420W)	Missense	Zou et al 2013[23]
HCM11.1	Mixed ancestry	Yes	MYBPC3	12	c.1000G>A (p.E334K)	Missense	Bahrudin et al 2008[24]
HCM14.1	European	No	MYH7	20	c.2167C>T (p.R723C)	Missense	Novel
HCM16.1	European	Yes	MYH7	9	c.746G>A (p.R249Q)	Missense	Zou et al 2013[23]
HCM21.1	Black African	Yes	MYBPC3	6	c.772G>A (p.E258K)	Missense	Andersen et al 2004[25]
HCM33.1	Mixed ancestry	Yes	MYBPC3	5	c.530G>A (p.R177H)	Missense	University of Stellenbosch thesis[26]
HCM34.1	Black African	Yes	MYH7	14	c.1357C>T (p.R453C)	Missense	Zou et al 2013[23]
HCM38.1	Black African	Yes	MYBPC3	15	c.1246G>A (p.G416S)	Missense	Tanjore et al 2008[27]

HCM, hypertrophic cardiomyopathy; *MYBPC3*, cardiac myosin-binding protein C; *MYH7*, cardiac β-myosin heavy chain.

The total number of variants in the 15 genes per HCM patients (regardless of whether it was disease-causing or not) ranged from 6 to 20 (mean: 12.8, SD 3.2; median 12). The patient who died had a higher number of variants (14.8±3.9) compared to survivors (12.5±3.1), although this difference was not statistically significant (P=0.47).

7.3.4. Outcome of HCM in South Africans

The mean duration of follow-up was 9.1 ± 3.4 years. Of the 43 patients studied, 8 died during the period of follow-up. The overall Kaplan-Meier survival estimate is shown in Fig. 7.1A; the cumulative proportion of patients who survived to 10 years was 74%. Complications of chronic heart failure, atrial fibrillation, stroke and evolution to dilated cardiomyopathy with systolic dysfunction were observed in 11 (25.6%), 8 (18.6%), 4 (9.3%), and 4 (9.3%), respectively. Therapeutic interventions including surgical myomectomy, alcohol septal ablation and orthotopic heart transplantation were performed on 3 (7.0%), 1 (2.3%) and 1 (2.3%) patients, respectively. At the last visit, 12 (27.9%) reported NYHA functional class III and IV performance status (Table 7.3). The most frequently prescribed drugs were beta-blockers and calcium channel blockers, used by 33 (76.7%) and 17 (39.5%) patients, respectively (Table 7.4).

Cox's proportional hazards regression showed that survival was predicted by NYHA functional class at last visit (P=0.026), but not by presence of a disease-causing mutation (P=0.474), as shown in Figures 7.1B and 7.1C, respectively. Survival in this cohort was similar to that of an age- and sex-matched general South African population (Figure 7.1D).

The presence of chronic heart failure (hazard ration (HR) 4.4, CI 1.0 – 18.3; p=0.044) and NYHA functional class at last visit (HR 6.2, CI 1.2 – 30.6; p=0.026) were found to be predictors of mortality on univariate regression analysis. On multivariable analysis, both chronic heart failure and NYHA functional class were not significant as predictors of mortality, as they may be proxies for LV ejection fraction (Table 7.5).

Table 7.3. Follow-up and outcome data

Mean duration of follow-up (years ± SD)	9.1 ± 3.4
Total number of mutations per person	12.8 ± (3.2)
Follow-up observation Regular Lost to follow-up	 37 (86.0) 6 (14.0)
Death	8 (18.6)
Chronic heart failure	11 (25.6)
ICD insertion	0 (0)
PPM insertion	0 (0)
CRT/Biventricular pacing	0 (0)
Loop recorder	6 (14.0)
Arrhythmia present No arrhythmia Atrial fibrillation Atrial flutter Ventricular tachycardia	 32 (74.4) 8 (18.6) 1 (2.3) 2 (4.7)
Myomectomy	3 (7.0)
Alcohol septal ablation	1 (2.3)
Evolution to DCM	4 (9.3)
Orthotopic heart transplantation	1 (2.3)
NYHA FC at last visit Class 1 and 2 Class 3 and 4	 31 (72.1) 12 (27.9)
Stroke	4 (9.3)

All values are number (percentage), unless otherwise stated.
ICD, implantable cardioverter-defibrillator; PPM, permanent pacemaker; CRT, cardiac resynchronisation therapy; DCM, dilated cardiomyopathy; NYHA, New York Heart Association functional classification for evaluation of severity of dyspnea.

Table 7.4. Medical therapy at follow-up

B-blocker	33 (76.7)
Calcium channel blocker	17 (39.5)
Warfarin	12 (27.9)
ACE-I or ARB	9 (20.9)
Furosemide	8 (18.6)
Aspirin	8 (18.6)
Disopyramide	4 (9.3)
Spironolactone	4 (9.3)
Amiodarone	3 (7.0)
Digoxin	2 (4.7)
Nitrates	1 (2.3)

ACE-I, angiotensin converting enzyme inhibitor; ARB, angiotensin receptor blocker; B-blocker, beta blocker.

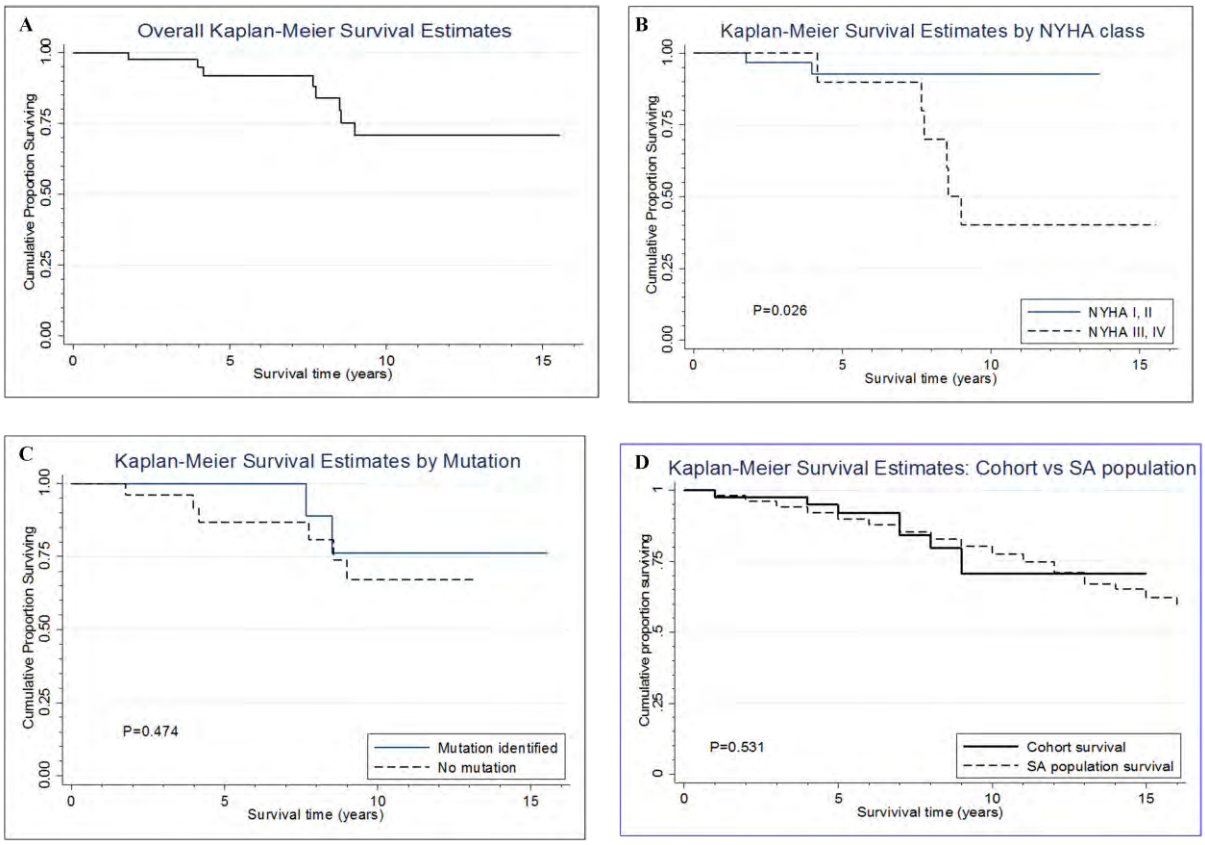

Figure 7.1. Kaplan-Meier survival estimates in hypertrophic cardiomyopathy

A. Kaplan-Meier plot showing the survival of HCM patients; B. Kaplan-Meier plot showing the survival of HCM patients when stratified by NYHA functional class; C. Kaplan-Meier plot showing the survival of HCM patients when stratified by presence or absence of HCM-causing mutation(s); and D. Kaplan-Meier plot showing the survival of HCM patients when compared to age-, sex- and race-matched members of the South African population

Table 7.5. Cox's proportional hazards regression model analysis of predictors of mortality in hypertrophic cardiomyopathy

Univariate Cox Regression		
	Hazard ratio (95% CI)	**P-value**
Age at diagnosis	1.0 (1.0 – 1.1)	0.561
Mutation positive	1.8 (0.4 – 8.9)	0.474
Sarcomeric mutations	1.3 (0.5 – 3.5)	0.585
Total number of mutations per person	1.12 (0.91 – 1.3)	0.412
IVS	1.6 (0.8 – 3.4)	0.169
LVEF	1.1 (1.0 – 1.2)	0.060
Family history of SCD	0.8 (0.1 – 6.6)	0.840
E/A ratio	2.0 (0.4 – 10.0)	0.370
Loop recorder	3.5 (0.8 – 14.6)	0.088
Chronic heart failure	4.4 (1.0 – 18.3)	0.044
NYHA functional class at last visit	6.2 (1.2 – 30.6)	0.026
Multivariate Cox regression		
	Hazard ratio (95% CI)	**P-value**
LVEF	1.1 (1.0 – 1.2)	0.100
Loop recorder	0.8 (0.1 – 5.3)	0.828
Chronic heart failure	1.6 (0.2 – 16.2)	0.684
NYHA functional class at last visit	4.2 (0.4 – 41.3)	0.218

E/A, ratio of early (E) to late (A) ventricular filling velocities on Doppler echocardiography; IVS, interventricular septal thickness in diastole; HCM, hypertrophic cardiomyopathy; LVEF, left ventricular ejection fraction; NYHA, New York Heart Association functional classification for severity of breathlessness; SCD, sudden cardiac death. In the univariate and multivariate regression analysis, NYHA was correlated as a binary variable (NYHA FC I-II vs. NYHA FC III-IV).

7.4 Discussion

To our knowledge this is the first prospective study of the clinical profile, spectrum of disease-causing gene mutations and outcome in HCM from the African continent, including black Africans. Age at onset of symptoms (38.5 ± 14.3 years), male preponderance (58%), and major symptoms were similar to those reported in North American, Middle Eastern, and Eastern series (Table 1).[166,170,171] Nearly 30% of patients bear mutations in the MYH7 and MYBPC3 genes which are the commonest genetic causes of HCM.[53] Whilst the annual mortality rate of 2.9% was high and the overall survival of 74% at 10 years is low compared to other series of patients with HCM,[166] the

survival rate was comparable to age- and sex-matched members of the South African population. Survival was predicted by New York Heart Association (NYHA) functional class at last visit.

We have found that HCM occurs predominantly in men, with a young age of onset, including black Africans; and with a positive family history of HCM in the majority. Fatigue, breathlessness and palpitations were the commonest symptoms. Atrial fibrillation was found in 9%, LVOT obstruction in 28%, and diastolic dysfunction in most. In a study of the natural history of HCM in non-hospitalised Americans, Maron and others found that 55% of patients were men and the mean age was 47 years, and cardiac symptoms were present in 63% of patients.[166] Similarly, in a study from Taiwan, Lee and colleagues found 52% HCM patients to be male, and that men had a younger age of onset of HCM compared to women;[170] in this study the prevalence of apical HCM was three times higher in men, and interestingly, men had lower prevalence of LVOT obstruction. Thirty-six percent of Taiwanese HCM patients had pulmonary oedema or paroxysmal atrial fibrillation. More recently, in the first report on the clinical characteristics of HCM in Saudi Arabia, Ahmed and coauthors found the population of HCM patients to be 71% male, and with a mean age of 42 years.[171] Dyspnoea and palpitations were the commonest symptoms, and LVOT obstruction was found in 28%.

To date, over 1400 mutations have been reported to cause HCM in genes encoding eight sarcomere proteins: beta-myosin heavy chain (MYH7), cardiac myosin-binding protein C (MYPBC3), cardiac troponin T (TNNT2), cardiac troponin I (TNNI3), cardiac actin (ACTC), alpha-tropomyosin (TPM1), essential light chain of myosin (MYL3) and regulatory light chain of myosin (MYL2).[53,172] Mutations in MYH7 and MYPBC3 occur most often, and account for approximately 50% of HCM cases,[173,174] while mutations in TNNT2, TNNI3, ACTC, TPM1, MYL3 and MYL2 collectively

account for less than 20% of HCM cases.[175] In our study, mutations in *MYH7* and *MYPBC3* were the commonest causes of HCM.

Moolman-Smook and colleagues have done pioneering work on the genetics of HCM in two South African subpopulations: those of European descent and those of mixed ancestry, and have previously reported on common HCM-causing mutations that arose independently and demonstrated clear founder effects in the South African population. These mutations included the MYH7 Ala797Thr (25% prevalence),[163] TNNT2 Arg92Trp (15%),[164] MYH7 Arg403Trp (5%),[162] MYH7 Arg717Gln and the MYH7 Glu499Lys[176] mutations, which collectively accounted for 47.5% of cases of HCM from the Eastern and Western Cape provinces of South Africa. To save money and to improve efficiency, a strategy was proposed to first screen for these 5 founder mutations before undertaking an extensive molecular genetic screening for other HCM mutations in South Africa.[165] However, in our study of 42 South African HCM patients, these founder mutations were absent.

The mutation yield of screening 15 sarcomeric and non-sarcomeric genes that are associated with HCM was relatively low in this study. Disease-causing mutations in any one of sarcomeric protein genes are found in up to two thirds of patients with HCM, and the yield of screening associated causal genes ranges from 40-70%.[53] The indications for molecular genetic testing in cardiomyopathy vary according to the yield of molecular testing, the cost of the molecular analyses, and impact of genetic testing on the medical management of the individual and the family. Given the relatively low yield of screening in this study, molecular genetic testing in Africans with HCM should probably not be carried out routinely as yet until studies of the full spectrum of causal mutations and of the impact of genetic testing on outcome are available.

In our study, the mean duration of follow-up was 9.1 years with an annual mortality rate of 2.9%; and complications included heart failure, atrial fibrillation, stroke and evolution to DCM. Myomectomy, alcohol septal ablation and heart transplantation were performed in a small number of patients; however no implantable-cardioverter defibrillators (ICDs) were used. The high rates of mortality observed in our study may reflect, in part, the higher mortality of the South African population as well as the skewed nature of tertiary centre experience with many symptomatic patients. In the US, HCM was found to have an annual mortality rate of 1.3%, and to be associated with stroke, atrial fibrillation, sudden cardiac death, congestive heart failure and need for heart transplantation.[166] In Taiwan, HCM was reported to have an annual mortality rate of 0.8%, and mortality to be predicted by LVOT obstruction, atrial fibrillation and female gender.[170] In Saudi Arabia, HCM had an annual mortality rate of 0.7%, with 5 ICDs inserted over 7 years of follow-up and a single patient progressing to end-stage dilated cardiomyopathy.[171]

This sub-study has a number of important limitations. First, the small sample size is a major weakness. This may account for the failure to detect the effect of known predictors of mortality in HCM, such as history of syncope and magnitude of LVH. Second, we screened for 15 genes that are commonly associated with HCM. However, there are several important HCM-causing mutations in other genes that were not included in our genetic panel, such as titin (TTN), myosin heavy chain gene (MYH6) and cardiac troponin C (TNNC). Therefore, there is a need for larger prospective studies of HCM in Africa that encompass all the important genetic causes of the disease.

7.5 Conclusions

We report on the first prospective investigation of the clinical characteristics, genetics, and outcome of HCM in Africans. We found HCM to occur most in men, and to have a young age of onset. Major symptoms and complications were similar to those reported in North American, Middle Eastern, and Asian studies. Known and novel disease-causing mutations were identified in the MYH7 and MYBPC3 genes with a lower yield of mutation screening of about 30% compared to the expected 40-70% elsewhere. The mortality in this contemporary African HCM series was, however, higher than reported elsewhere, although comparable to age- and sex-matched members of the South African population. Survival was predicted by New York Heart Association (NYHA) functional class at last visit.

Chapter 8

Genotype: phenotype correlations in hypertrophic cardiomyopathy patients with the three known South African founder mutations: a cardiovascular magnetic resonance study

8.1 Introduction

Hypertrophic cardiomyopathy (HCM) is an inherited disease of the sarcomere characterised by ventricular myocardial hypertrophy, myocyte disarray and cardiac fibrosis, in the context of a non-dilated LV,[8] and in the absence of increased external load.[76,177] HCM is caused by over 1400 mutations found in at least 20 causal genes that encode sarcomeric and calcium-handling proteins, as well as in genes that encode other proteins (e.g. Z-disc proteins) that interact with sarcomeric proteins.[173,174] Furthermore, mitochondrial mutations, lysosomal storage diseases, as well as infiltrative cardiomyopathies may be HCM phenocopies;[175] however the molecular aetiologies of these disorders are distinct from HCM.

The known HCM-causing genes are useful for (1) effective gene-based diagnosis of HCM, (2) accurate assessment of disease risk in family members, and (3) providing insight into likely clinical course of HCM in affected individuals.[173] Several studies have attempted to correlate the clinical features and course of HCM to a specific causal mutation; and such genotype: phenotype studies have implied that the causal allele is responsible, in part, for the variability observed in HCM

phenotype. Specific mutations have been linked with extensive left ventricular hypertrophy (LVH), mid-cavity obstruction, apical hypertrophy, age of onset, risk of sudden cardiac death (SCD), incidence of congestive cardiac failure, risk of progression to cardiac dilation and conduction disease.[178-187] However, none of these mutations have been exclusively associated with the proposed HCM phenotypes. Furthermore, the pathophysiological pathway between genetic mutation and resulting clinical HCM phenotype is incompletely understood.

There is significant overlap between the clinical phenotype of unexplained LVH and HCM-causing mutations, making the utility of clinical findings to accurately predict pathophysiologic involvement of a particular gene difficult. Moreover, genotypic: phenotypic correlations have been limited by small numbers of genotyped HCM patients, small sizes of families included in studies, differing age distributions of mutation-carriers within families, low individual frequency of HCM-causing gene mutations, variable clinical expression within and between families, and poor understanding of the genetic heterogeneity that modifiers such as background genotypes, gender and environment have on phenotypic expression.[188-190] Despite all these complexities, it is generally recognised that the clinical course of HCM tends to be more severe in those with an identified causative genetic mutation and in those with multiple HCM-causing mutations.[191]

Extensive genetic studies in South African HCM patients have identified three unique HCM-founder alleles: the A797T mutation and the R403W mutation in the beta-myosin heavy chain (*MHY7*) gene, and the R92W mutation in the cardiac troponin T (*TNNT2*) gene.[162-165,192] Haplotype analysis has confirmed that these mutations are identical-by-descent, indicating that individuals bearing the same mutation are ancestrally related.[165] The purpose of this sub-study was to investigate whether there were any correlations between genotype and phenotypic manifestations, based on cardiovascular magnetic resonance (CMR) features.

8.2 Methods

8.2.1. Study design

The study was designed as a cross-sectional study of the relation of CMR phenotypic characteristics to genotype in a cohort of consecutive patients diagnosed with HCM, due to the three known South African HCM-founder alleles ($A797T_{MYH7}$; $R403W_{MHY7}$; and $R92W_{TNNT2}$) at Tygerberg Hospital in Cape Town, South Africa. CMR imaging was carried out at Groote Schuur Hospital, Cape Town, South Africa.

8.2.2. Study population and definitions

151 HCM mutation carriers (MCs) were successfully enrolled into the study. Seven patients had incomplete CMR studies. Another 11 patients had non-diagnostic CMR studies either due to presence of motion artefacts, mistriggering consequent to arrhythmias, or suboptimal late gadolinium enhancement (LGE) imaging. Two MCs had CMR evidence of dilated cardiomyopathy, and were subsequently also excluded from the analysis (Figure 8.1). 131 HCM MCs were included in the analysis (mean age 39.83 ± 16.2 years; 58% male). A sensitivity analysis indicated that the demographic profile of the 20 patients excluded from the analysis was similar to that of the 131 patients analysed, and thus unlikely to have biased the analysis.

The diagnosis of HCM was based on the presence of a hypertrophied (maximum end-diastolic wall thickness ≥ 14mm), non-dilated LV in the absence of other diseases capable of producing the degree of observed hypertrophy. Arterial hypertension was defined as a blood pressure greater than 140/90 mmHg. Obesity was defined as a body mass index greater than 30 kg/m^2.

8.2.3. CMR measurements and data collection

Subjects were scanned in a 1.5T MRI scanner (Symphony, Siemens Medical Systems, Erlangen, Germany) using a cardiac phased array coil. Initially, scout images were acquired with 2- and 4-

chamber views, and were used to prescribe short-axis planes from base to apex for *cine* imaging. Imaging with steady-state free precession (SSFP), with segmented acquisition of k-space lines, was applied for *cine* imaging, with breath-holding by the patient for ± 6-10 seconds (~15 cardiac cycle lengths). The LV systolic function and mass were imaged with *cine* MRI along contiguous short-axis slices (typically 1.4x1.4 mm/pixel and 7 mm slice thickness, with a 3 mm slice gap). The sequence parameters were as follows: repetition time (TR) 35 ms; echo time (TE) 1.5 ms; receiver bandwidth 930Hz/pixel; 256 readout points with oversampling; 165 phase encodings; 65º flip angle; a field of view with dimensions of 256mm in the phase encoding direction and 340mm in the read-out direction; and ≥15 phases per R-R interval. LGE imaging was performed with an inversion recovery method, after a dose of 0.2 mmol/kg of gadolinium-DTPA (gadodiamide, Omniscan®, General Electric Healthcare) with the inversion time optimized iteratively to maximally null the myocardium.

8.2.4. CMR data analysis

i. LV structure and function

Cine CMR images were analysed offline with a validated image-processing software package (ARGUS; Siemens Medical Systems, Erlangen, Germany). End-diastolic and end-systolic images were identified with electrocardiographic (ECG) R-wave gating, and LV volume assessment. Contours of the endocardial and epicardial borders were manually delineated by two experienced observers, who were blinded to the genotypic information, from base to apex in end-diastole. Only endocardial contours were drawn in end-systole. LV end-systolic (LVESV) and end-diastolic (LVEDV) volumes were used to calculate stroke volume (SV) and ejection fraction (EF) – (EF = SV/EDV). Myocardial mass was calculated by subtracting the endocardial volume from the epicardial volume, based on prior knowledge of myocardial specific gravity (1.05 g/cm3). Left atrial diameter was measured in the LV outflow tract (3-chamber) view.

ii. Late gadolinium enhancement

Images were evaluated in a blinded fashion, for the presence or absence, pattern (focal, confluent or mixed), amount and regional distribution of LGE areas by a two observers with greater than 4 years of CMR experience.

8.2.5. Ethical considerations

The study was designed in keeping with the principles of the Helsinki Declaration. The study was approved by the Human Research Ethics Committees of Stellenbosch University and the University of Cape Town. All participants gave informed, written consent to participate in the study.

8.2.6. Statistical analysis

Normality of data was tested using the Kolmogorov-Smirnov test. Normally distributed data are presented as mean ± standard deviation (SD) or, where highly skewed, as median (interquartile range - IQR); categorical data are presented as numbers (percentages). The chi-square test or Fischer's exact test was used to compare dichotomous data. The unpaired Student t-test (when normally distributed) or Mann- Whitney U test (for non-parametric data) was used to compare continuous variables. Bivariate correlations were assessed using the Pearson's (R) or the Spearman's (Rs) coefficient, as appropriate. Regression analysis used to determine relationship between LVH and LGE. All statistical tests were two-tailed and a p-value of less than 0.05 was considered statistically significant. All analysis was performed using SPSS version 20 (IBM, Armonk, New York, USA).

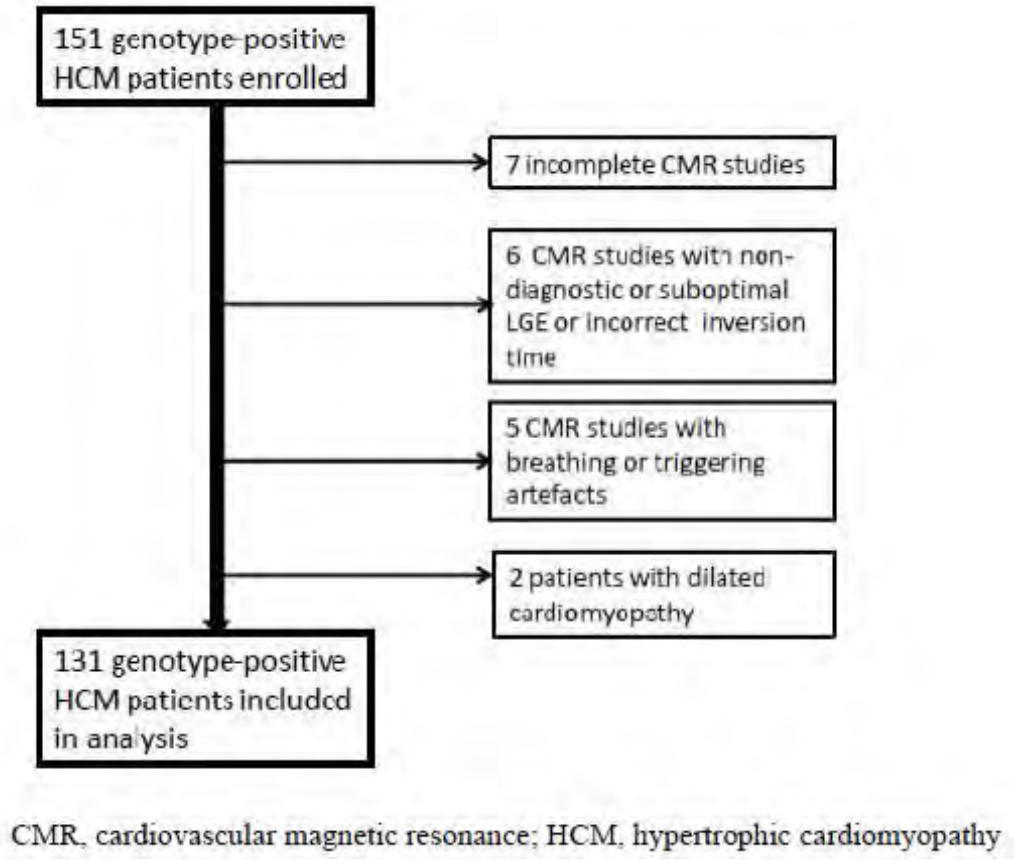

Figure 8.1. HCM patients enrolled into the study

8.3 Results

8.3.1. Baseline characteristics

The demographic details of the HCM MCs included in the analysis are shown in Table 8.1.

There were no differences in age, sex or co-morbid status when study participants were stratified by genotype. The prevalence of obesity and hypertension were low, likely reflecting the relatively young age of the study participants.

Table 8.1. Baseline characteristics of HCM patients stratified by genotype

Characteristic	$A797T_{MYH7}$ N=58	$R403W_{MYH7}$ N=30	$R92W_{TNNT2}$ N=43	P Value
Age (years ± SD)	43.3 ± 16.4	36.6 ± 14.3	37.4 ± 16.5	0.093
Male sex (%)	36 (62.1)	20 (66.7)	20 (46.5)	0.161
Hypertension (%)	7 (12.1)	3 (10.0)	7 (16.3)	0.707
Systolic BP (mmHg ± SD)	123.3 ± 12.3	120.0 ± 11.7	119.1 ± 14.6	0.301
Diastolic BP (mmHg ± SD)	78.3 ± 9.6	75.0 ± 11.4	73.9 ± 11.6	0.112
Obesity (%)	9 (15.5)	7 (23.3)	11 (25.6)	0.426
BMI (kg/m^2 ± SD)	25.8 ± 4.4	26.1 ± 3.5	25.1 ± 5.8	0.591

$A797T_{MYH7}$, the A797T mutation in the beta-myosin heavy chain gene; $R403W_{MYH7}$, the R403W mutation in the beta-myosin heavy chain gene; $R92W_{TNNT2}$, the R92W mutation in the cardiac troponin T gene; BP, blood pressure; BMI, body mass index

8.3.2. CMR findings: LV structure and function

Global systolic function was normal in all 3 HCM groups studied, as shown in Table 8.2. While within normal limits, there was a small but statistically significant increase in LVEF of $R92W_{TNNT2}$ MCs when compared to $A797T_{MYH7}$ and $R403W_{MYH7}$ MCs, (73.4 ± 6.3 vs. 67.2 ± 6.9 and 70.7 ± 6.9, respectively) p=0.013. The $R403W_{MYH7}$ MCs had greater frequency of inferior wall LVH compared to $A797T_{MYH7}$ and $R92W_{TNNT2}$ MCs, with maximal LVH occurring in the interventricular septum (p=0.043). There were also differences noted in the pattern of LVH between participants with different mutations. For instance, the $R92W_{TNNT2}$ MCs had higher frequency of combined septal and apical hypertrophy; the $A797T_{MYH7}$ MCs had more cases of isolated apical HCM; and the $R403W_{MYH7}$ MCs had greater prevalence of asymmetric septal hypertrophy (p=0.006). Examples of patterns of LVH in HCM are depicted in Figure 8.2. There were no differences in the prevalence of left ventricular outflow tract (LVOT) obstruction between the different genotypes (p=0133). Also, there were no differences in left atrial (LA) size between the different groups (p=0.881).

8.3.3. CMR findings: regional fibrosis assessed by LGE

The presence of LGE, assessed qualitatively, was higher in R403W$_{MYH7}$ MCs compared to A797T$_{MYH7}$ and R92W$_{TNNT2}$ MCs (p=0.023), shown in Table 8.2. Focal LGE was found more commonly in A797T$_{MYH7}$ and R403W$_{MYH7}$ MCs, while the R92W$_{TNNT2}$ MCs commonly had confluent or mixed patterns of LGE (p=0.001), Figure 8.3.

8.3.4. Genotype correlations with CMR covariates of IVS thickness and LGE

In A797T$_{MYH7}$ MCs, interventricular septal (IVS) thickness correlated with male sex (R_S 0.333; p=0.011), left ventricular end-diastolic volume [LVEDV] (R_S -0.638; p<0.001), LV mass (R_S 0.642; p<0.001), LA size (R_S 0.602; p<0.001), LVOT obstruction (R_S 0.509; p<0.001), LGE presence (R_S 0.756; p<0.001) and presence of mitral regurgitation (R_S 0.411; p=0.001), as depicted in Table 8.3. In R403W$_{MYH7}$ MCs, IVS thickness was found to correlate with age (R_S 0.511; p=0.004), LVEDV (R_S -0.393; p=0.032), LV mass (R_S 0.503; p=0.005) and LGE presence (R_S 0.363; p=0.049). In R92W$_{TNNT2}$ MCs, IVS thickness correlated with age (R_S 0.563; p<0.001), LVEDV (R_S -0.461; p=0.002), LV mass (R_S 0.571; p<0.001), LA size (R_S 0.591; p=0.001), LVOT obstruction (R_S 0.330; p=0.031), LGE presence (R_S 0.656; p<0.001) and presence of mitral regurgitation (R_S 0.538; p<0.001).

In A797T$_{MYH7}$ MCs, presence of LGE correlated with age (R_S 0.281; p=0.033), LVEDV (R_S -0.551; p<0.001), LV mass (R_S 0.373; p=0.004), IVS thickness (R_S 0.756; p<0.001), LA size (R_S 0.557; p<0.001) and presence of mitral regurgitation (R_S 0.359; p=0.006), shown in Table 8.4. In R403W$_{MYH7}$ MCs, presence of LGE correlated with LA size (R_S 0.393; p=0.032) and IVS thickness (R_S 0.363; p=0.049). In R92W$_{TNNT2}$ MCs, LGE presence correlated with LVEDV (R_S -0.323; p=0.035), LA size (R_S 0.379; p=0.012) and IVS thickness (R_S 0.656; p<0.001).

Table 8.2. CMR findings in HCM patients stratified by genotype

Characteristic	A797T$_{MYH7}$ N=58	R403W$_{MYH7}$ N=30	R92W$_{TNNT2}$ N=43	P Value
LVEDV indexed to BSA (ml/m^2 ± SD)	62.4 ± 14.7	64.3 ± 11.6	68.1 ± 12.1	0.105
LVEF	67.2 ± 6.9	70.7 ± 6.9	73.4 ± 6.3	0.013
LV mass indexed to BSA (g/m^2 ± SD)	61.2 ± 14.5	64.8 ± 12.1	61.3 ± 17.2	0.521
LA size (mm ± SD)	34.2 ± 7.6	33.8 ± 6.3	33.5 ± 6.4	0.881
IVS in end-diastole (mm ± SD)	17.3 ± 7.6	17.7 ± 6.1	15.0 ± 6.1	0.166
Position of maximal LV thickness (%)				
Septum	54 (93.1)	25 (83.3)	40 (93.0)	
Anterior wall	0 (0)	0 (0)	2 (4.7)	
Inferior wall	4 (6.9)	5 (16.7)	1 (2.3)	0.043
Type of HCM (%)				
HCM with basal asymmetrical septal bulge	6 (10.3)	7 (23.3)	3 (9.3)	
Mid-ventricular asymmetrical septal hypertrophy (including mid-cavitary HCM)	30 (51.7)	15 (50.0)	13 (30.2)	
Apical HCM	3 (5.2)	0 (0)	0 (0)	
HCM with concentric left ventricular hypertrophy	1 (1.7)	2 (6.6)	0 (0)	
Mass-like HCM	0 (0)	0 (0)	0 (0)	
HCM with non-contiguous areas of left ventricular hypertrophy (including combined septal and apical HCM)	0 (0)	2 (6.6)	7 (16.3)	
'Burnt-out'' HCM	1 (1.7)	0 (0)	0 (0)	
Normal CMR (no HCM features)	17 (29.3)	4 (13.3)	20 (46.5)	0.006
Presence of LVOT obstruction (%)	6 (10.3)	2 (6.7)	4 (9.3)	0.133
Presence of MR (%)	12 (20.7)	6 (20.0)	13 (30.2)	0.467
Presence of LV myocardial crypts (%)	5 (8.6)	6 (20.0)	6 (14.0)	0.276
RV hypertrophy (%)	2 (3.4)	0 (0)	1 (2.3)	0.689
Apical aneurysm (%)	5 (8.6)	3 (10.0)	5 (11.6)	0.930
Genotype positive, phenotype positive (%)	41 (70.7)	26 (86.7)	23 (53.5)	0.010
Presence of LGE (%)	33 (56.9)	23 (76.7)	25 (58.1)	0.023
Pattern of LGE (%)				
Focal	19 (32.8)	12 (40.6)	5 (11.6)	
Confluent	14 (24.1)	11 (36.7)	14 (32.6)	
Mixed	0 (0)	0 (0)	6 (14.0)	0.001

LVEDV, left ventricular end-diastolic volume; BSA, body surface area; LVEF, left ventricular ejection fraction; LA, left atrium; IVS, interventricular septum; LVOT, left ventricular outflow tract obstruction; MR, mitral regurgitation; RV, right ventricle; LGE, late gadolinium enhancement

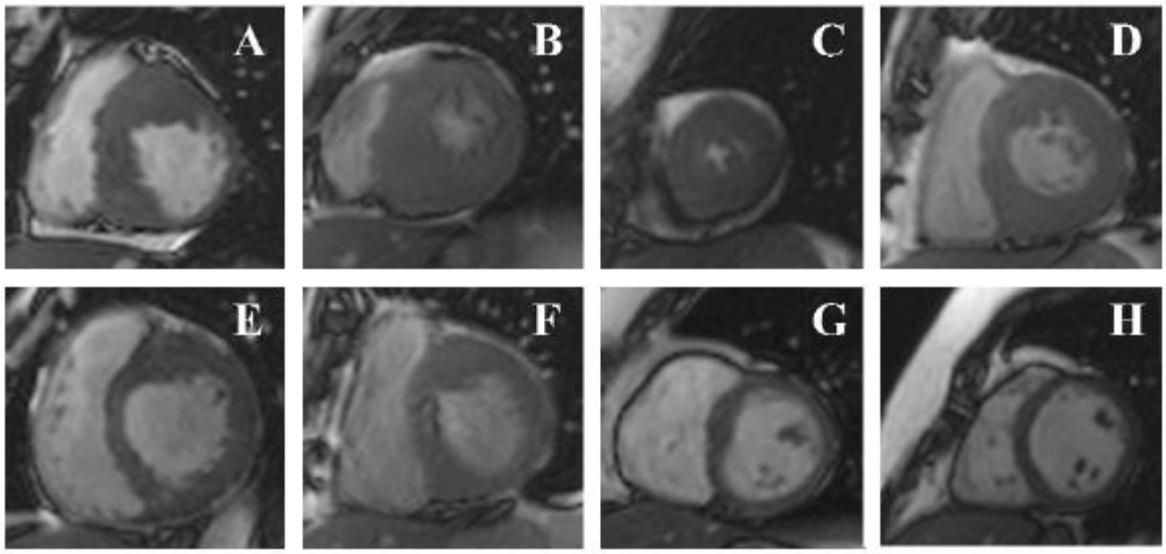

Figure 8.2. Patterns of LV hypertrophy found in HCM, demonstrated in short-axis images in end-diastole
A, HCM with focal basal septum and/or anterior bulge; B, mid-ventricular asymmetrical septal left ventricular hypertrophy (including mid-cavitary HCM); C, apical HCM (short-axis apical diastolic slice showing apical hypertrophy); D, HCM with concentric left ventricular hypertrophy; E, mass-like HCM (in this case, showing focal thickening in the anterior myocardium); F, HCM with non-contiguous areas of left ventricular hypertrophy (including septal and inferior wall LVH, septal and apical LVH, or septal and lateral wall LVH); G, burnt-out HCM (with thinned and fibrotic myocardium and decreased LV systolic function); and H, genotype positive, phenotype negative HCM (with a normal CMR appearance)

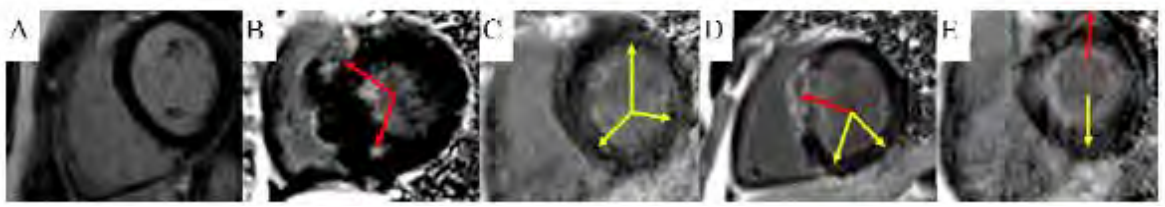

Figure 8.3. Patterns of LGE in HCM
A, LGE in a healthy control; B, HCM patient with focal enhancement (shown in red arrows) in the superior and inferior LV/RV junctions; C, HCM patient with confluent (diffuse and patchy) enhancement throughout the myocardium (indicated in yellow arrows); D, HCM patient showing a mixed pattern of enhancement with focal enhancement in the hypertrophied septum (red arrow) an confluent mid-wall enhancement in the inferolateral wall; and E, another HCM patient with a mixed pattern, with a small area of focal fibrosis (red arrow) in the anterior wall and mostly confluent fibrosis (yellow arrow) in the inferolateral wall and septum

Table 8.3. Genotype correlations with CMR covariates of interventricular septal thickness

Characteristic	$A797T_{MYH7}$ N=58		$R403W_{MYH7}$ N=30		$R92W_{TNNT2}$ N=43	
	R_S	*P value*	R_S	*P value*	R_S	*P value*
Male sex	0.333	0.011	0.239	0.203	0.267	0.084
Advancing age	0.235	0.076	0.511	0.004	0.563	<0.001
$LVEDV_{indexed\ to\ BSA}$	-0.638	<0.001	-0.393	0.032	-0.461	0.002
$LV\ Mass_{indexed\ to\ BSA}$	0.642	<0001	0.503	0.005	0.571	<0.001
LA size	0.602	<0.001	0.320	0.085	0.519	<0.001
LVOT obstruction	0.509	<0.001	0.132	0.486	0.330	0.031
Presence of LGE	0.756	<0.001	0.363	0.049	0.656	<0.001
Presence of MR	0.411	0.001	-0.029	0.879	0.538	<0.001

R_S, Spearman bivariate correlation; LV, left ventricle; LVEDV, left ventricular end-diastolic volume; LA, left atrium; LVOT, left ventricular outflow tract; LGE, late gadolinium enhancement; MR, mitral regurgitation

Table 8.4. Genotype correlations with CMR covariates of LGE

Characteristic	$A797T_{MYH7}$ N=58		$R403W_{MYH7}$ N=30		$R92W_{TNNT2}$ N=43	
	R_S	*P value*	R_S	*P value*	R_S	*P value*
Age	0.281	0.033	0.109	0.565	0.504	<0.001
$LVEDV_{indexed\ to\ BSA}$	-0.551	<0.001	-0.296	0.112	-0.323	0.035
$LV\ Mass_{indexed\ to\ BSA}$	0.373	0.004	0.123	0.518	0.247	0.110
IVS thickness	0.756	<0.001	0.363	0.049	0.656	<0.001
LA size	0.557	<0.001	0.393	0.032	0.379	0.012
LVOT obstruction	0.129	0.411	0.169	0.373	0.271	0.078
Presence of MR	0.359	0.006	0.079	0.679	0.350	0.201

R_S, Spearman bivariate correlation; LV, left ventricle; LVEDV, left ventricular end-diastolic volume; LA, left atrium; LVOT, left ventricular outflow tract; LGE late gadolinium enhancement; MR, mitral regurgitation

8.3.5. Predictors of LGE presence in HCM

On multivariate logistic regression analysis, LV mass was the only independent predictor of

LGE (OR 1.73 [1.35 – 8.08]; p=0.035), as shown in Table 8.5.

Table 8.5. Logistic regression analysis of predictors of LGE in HCM

Characteristic	Univariate logistic regression analysis	
	Odds ratio (95% CI)	*P Value*
Age	1.024 (1.000–1.858)	0.310
$LVEDV_{indexed\ to\ BSA}$	-2.330 (-1.209—2.822)	0.024
LVEF	0.908 (0.653–1.002)	0.321
IVS thickness	2.972 (1.06 –3.408)	0.004
LA size	3.604 (1.208–4.709)	0.001
$LV\ Mass_{indexed\ to\ BSA}$	2.493 (1.573–12.628)	0.008
	Multivariate logistic regression analysis	
	Odds ratio (95% CI)	*P Value*
Age	1.067 (1.008–1.137)	0.647
$LVEDV_{indexed\ to\ BSA}$	-0.662 (-0.335—0.903)	0.350
LVEF	0.824 (0.587–0.913)	0.418
IVS thickness	1.031 (1.001–1.209)	0.067
LA size	1.243 (0.907–1.533)	0.109
$LV\ Mass_{indexed\ to\ BSA}$	1.730 (1.346–8.075)	0.035

LV, left ventricle; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LA, left atrium

8.4 Discussion

The key findings of the first detailed report on the CMR phenotype in a cohort of HCM mutation carriers with the three known South African HCM-founder alleles were: (i) global systolic function was within the normal range in all 3 groups, however, the $R92W_{TNNT2}$ MCs had increased LVEF in comparison to $A797T_{MYH7}$ and $R403W_{MYH7}$ MCs; (ii) $R403W_{MYH7}$ MCs had greater frequency of inferior wall and basal septal LVH and there were different patterns of LVH observed in MCs with different genotypes; (iii) LGE was more common in $R403W_{MYH7}$ MCs, with focal LGE more prevalent in $A797T_{MYH7}$ and $R403W_{MYH7}$ MCs, and $R92W_{TNNT2}$ MCs more frequently having confluent or mixed patterns of LGE; (iv) unique CMR covariates were found to correlate differently

with genotype; and (v) LV mass was the only independent predictor of LGE presence, with an odds ratio of 1.73. This study contributes to our evolving understanding of genetic influence on phenotypic expression in HCM patients with the three unique South African HCM-founder mutations and suggests that CMR may be a useful tool in distinguishing variable phenotypic expression in HCM MCs with differing genotypes.

On echocardiography, in South African HCM founder MCs with apparently mild disease, there was a positive correlation between age and IVS thickness for both the $R403W_{MYH7}$ and $R92W_{TNNT2}$ mutations.[193] In the same study, the period between development of LVH coincided with the years in which most SCD events occurred in male $R92W_{TNNT2}$ MCs.[193] We also demonstrate that in all 3 groups of South African HCM mutations, IVS thickness correlated negatively with LVEDV and positively with LV mass and presence of LGE. Similarly, we also find a positive correlation between age and IVS thickness in $R403W_{MYH7}$ and $R92W_{TNNT2}$ MCs. In the $A797T_{MYH7}$ and $R403W_{MYH7}$ MCs, there was a correlation between IVS thickness and increasing LA size. Furthermore, in the $A797T_{MYH7}$ and $R92W_{TNNT2}$ MCs, IVS thickness correlated with presence of LVOT obstruction and mitral regurgitation. Greater CMR-defined LV mass and IVS thickness are associated with less favourable clinical outcome, likely reflecting the strong association of LVH with LVOT obstruction, SCD and advanced heart failure.[194,195]

Regional myocardial fibrosis or scarring detected by LGE on CMR occurs in approximately 60% (range 30-90%) of HCM patients.[195-197] While there are no LGE characteristics that are specific for HCM,[198] the prognostic significance of LGE in HCM is high, as LGE is often associated with SCD, systolic dysfunction, and non-sustained ventricular tachycardia.[199-202] We show that in all 3 groups of South African HCM mutations, LGE correlated with LA size and IVS thickness. In the $A797T_{MYH7}$ and $R92W_{TNNT2}$ MCs, LGE also correlated with age and LVEDV. The relationship of LGE to LVEF remains controversial, as some investigators have found no relationship,[203] while

others have shown that percent of LV involvement by LGE inversely correlates with LVEF.[204,205] In this study, most patients has a normal ejection fraction. However, it is important to note that the degree of fibrosis in HCM has not been shown to be a predictor of major cardiovascular events on multivariate analysis,[202] likely due to small numbers of study subjects with events, and larger, multi-centre longitudinal studies are on-going for this assessment.

On multivariate analysis, we show that presence of LGE is associated with LV mass. Similarly, a significant but modest relationship has been demonstrated between LVH and increased LV mass and LGE.[204] Patients with LGE have greater LV mass index than patients without LGE. [201,205] Furthermore, on an individual level, a relationship is evident between segmental LV wall thickness and LGE.[204,205] Another comparison of echocardiographic structural and functional parameters between prehypertrophic South African MCs and non-MC family members revealed that (1) the $R92W_{TNNT2}$ mutation correlated with a relative increase in systolic functional parameters; (2) the $A797T_{MYH7}$ mutation correlated with reduced diastolic dysfunction; and (3) the $R403W_{MYH7}$ mutation correlated with both impaired systolic and diastolic dysfunction.[206] In this study, overall systolic function was normal in all 3 genotypic groups. However, we also found that $R92W_{TNNT2}$ MCs had a relative increase in global systolic function when compared with $A797T_{MYH7}$ and $R403W_{MYH7}$ MCs.

Our sub-study has several limitations. First, while there are several structural and functional genotypic correlates noted, there is significant overlap between these phenotypes. Second, the presence of correlation does not equate to causation. Nevertheless, the study was not designed to assess for causal links between genotype and phenotype. Third, it would have been interesting to assess the relationship of the CMR parameters with clinical outcomes in this unique cohort of HCM MCs, though this was not the purpose of this study. However despite these limitations, we

succeeded in achieving our primary objective of investigating genotype: phenotype correlations in HCM MCs due the South Africa founder alleles.

8.5 Conclusions

In this comprehensive report on genotype correlations with CMR phenotype, in a cohort of HCM MCs with the three known South African HCM-founder alleles, we show differences in pattern of LVH, and in frequency and pattern of LGE when comparing different genotypes. Furthermore, we also show that presence of LGE is related to LV mass index. Moreover, this study suggests that CMR is capable of distinguishing variable phenotypic expression in HCM MCs with differing genotypes, and contributes to our evolving understanding of the genetic influence on phenotypic expression in HCM patients with the three unique South African HCM-founder mutations.

Chapter 9

Conclusions

While little is known about the mechanisms, clinical characteristics, natural history and outcomes of cardiomyopathy amongst Africans, this series of studies aimed to broaden our understanding of cardiomyopathies among Africans and to investigate the clinical features and outcomes of both familial and non-familial cardiomyopathies. We hypothesise that cardiomyopathies are a caused by familial/genetic and non-familial/non-genetic factors, and that disease onset, clinical and pathological features, disease progression and severity, and disease prognosis vary based on familial/genetic and non-familial/non-genetic aetiology.. It is our belief that the series of sub-studies discussed above, indeed support this hypothesis. A major limitation of this work is that molecular genetics were not available for all cardiomyopathy patients studied at the completion of this thesis. However, these analyses are ongoing, and it is our belief that they will provide further evidence to support our contention of genetic underpinnings of cardiomyopathy.

The sarcomere has long been recognised as the principal contractile unit of striated muscle. A recent publication has supported our hypothesis, showing that mutations in genes encoding sarcomeric proteins are responsible for a range of diseases including hypertrophic, dilated and restrictive cardiomyopathies and ventricular non-compaction.[207] The authors critically appraise the evidence in this area and conclude that the downstream molecular pathways leading to these heterogeneous phenotypes include changes in acto-myosin cross-bridge kinetics, altered

mechanosensation, disturbed calcium sensitivity, deregulated signalling pathways, inefficient myocardial energetics, myocardial ischaemia and myocardial fibrosis.[208-212] Elucidation of the genetic causes of cardiomyopathy has helped in understanding the structure and function of the sarcomere and a more detailed knowledge of the sarcomere and its associated proteins has suggested additional gene candidates.

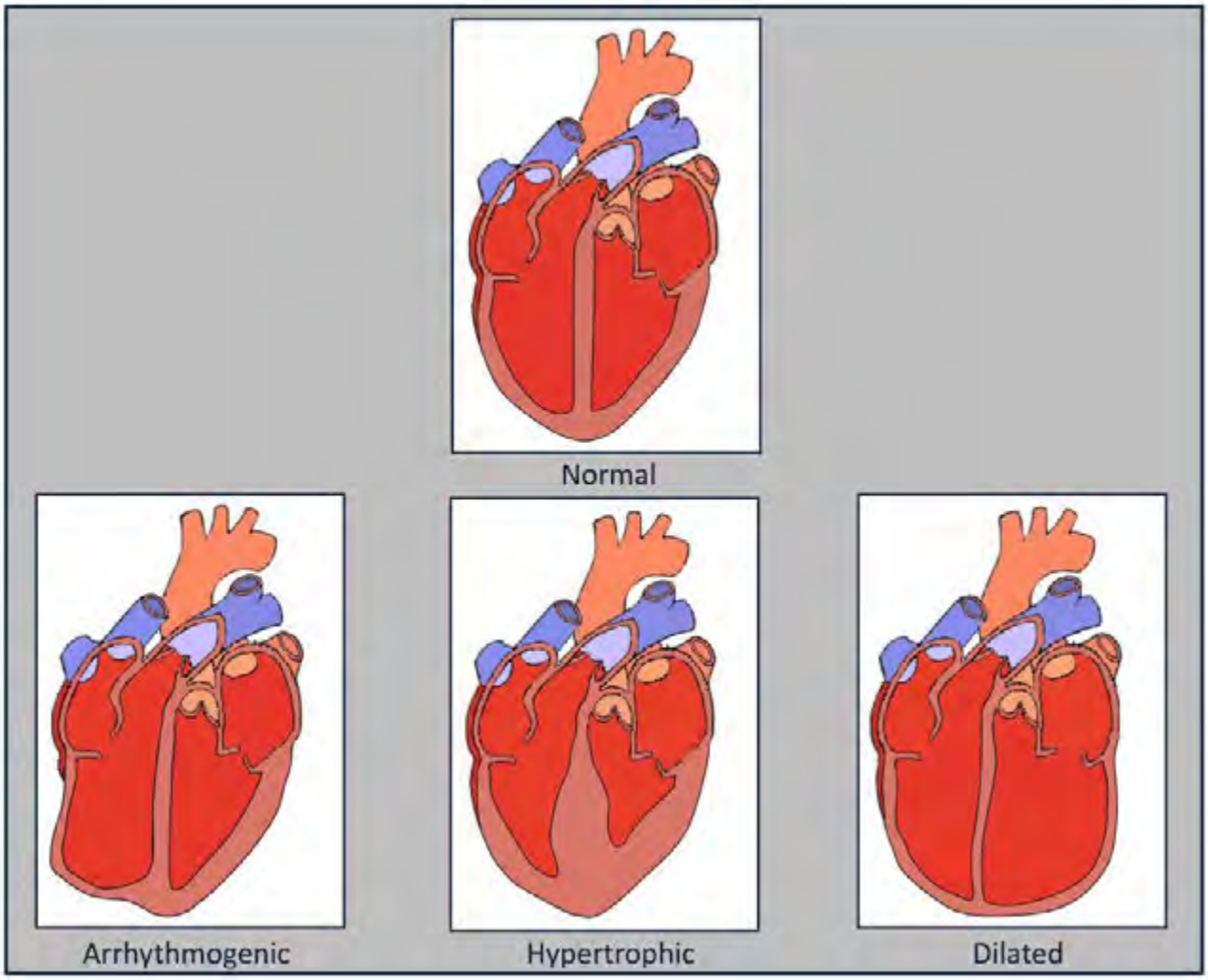

Figure 9.1. Schematic showing that sarcomeric mutations can lead to variable phenotypic expression in the cardiomyopathies
From Lopes LR, Elliott PM. A straightforward guide to the sarcomeric basis of cardiomyopathies. *Heart* 2014;100:1916-1923.

Table 9.1. Summary of the sarcomeric and associated proteins, encoded genes, interactions, associated disease phenotypes and respective prevalence

Gene	OMIM	Protein	Interactions	DCM	HCM	RCM	LVNC
Sarcomere contractile proteins							
MYH7	160760 192600	Myosin heavy chain, cardiac muscle β isoform	Myosin-binding protein C, myosin light chains, actin, titin, myomesin	X (4%)	X (20%)	X	X
MYH6	160710	Myosin heavy chain, cardiac muscle α isoform	Myosin-binding protein C, myosin light chains, actin, titin	X (4%)	X		
MYL2	160781 608758	Myosin regulatory light chain 2, ventricular/ cardiac muscle isoform	Myosin heavy chain		X (<1%)		
MYL3	160790 608751	Myosin light polypeptide 3	Myosin heavy chain		X (<1%)	X	
MYBPC3	600958	Myosin-binding protein C, cardiac type	Myosin heavy chain, titin, actin	X (2%)	X (20%)		X
TNNT2	115195	Troponin T, cardiac muscle	Troponin I, tropomyosin	X (3%)	X (2.5%)	X	X
TNNI3	191044	Troponin I, cardiac muscle	Actin, troponin C, troponin T	X (<1%)	X (2.5%)	X	
TNNC1	191040	Troponin C, slow skeletal and cardiac muscles	Troponin I	X (<1%)	X		
TPM1	115196 191010	Tropomyosin 1 α chain	Troponin T, actin	X (<1%)	X (1%)		X
ACTC1	102540	Actin, α cardiac muscle 1	Myosin heavy chain, tropomyosin, troponin I, myosin-binding protein C, α-actinin 2	X (<1%)	X (<1%)	X	X
Sarcomeric cytoskeleton							
TTN	188840	Titin	Multiple (see text)	X (18–25%)	X		
ACTN2	102573	α-actinin 2	Actin	X (1%)	X		
CSRP3	600824	Muscle LIM protein	α-actinin, calcineurin, telethonin	X (<1%)	X		
TCAP	604488	Titin-cap or telethonin	Myozenin-2, myostatin	X (1%)	X		
ILK	602366	Integrin-linked kinase	Muscle LIM protein	X (<1%)			
MYOZ2	606602	Myozenin	Telethonin		X		
FHL1	300163	Four-and-a-half-LIM domains 1					
LDB3	605906	LIM domain-binding protein 3/Cypher/ZASP	α-actinin, PKC	X (1%)	X		X
PDLIM3	605889	PDZ LIM domain protein 3		X (<1%)			
BAG3	603883	BCL2-associated athanogene 3		X			
DES	125660	Desmin		X (<1%)		X	
CRYAB	123590	α B crystalin	Desmin, vimentin, actin	X (< 1%)		X	
NEBL	605491	Nebullete	Actin				
MYPN	608517	Myopalladin	α-actinin, titin, nebulette, cardiac ankyrin repeat protein	X (3%–4%)			
NEXN	613121	Nexilin		X (<1%)	X		
ANKRD1	609599	Cardiac ankyrin repeat protein	Myopalladin/titin	X (2%)	X		

For HCM: only percentages for the strongest established genes are shown. X means that mutations in the gene have been associated with the phenotype.
DCM, dilated cardiomyopathy; HCM, hypertrophic cardiomyopathy; OMIM, online mendelian inheritance in man; LVNC, left ventricular non-compaction; PKA - protein kinase A; RCM, restrictive cardiomyopathy.

From Lopes LR, Elliott PM. A straightforward guide to the sarcomeric basis of cardiomyopathies. *Heart* 2014;100:1916-1923.

In this study, two cohorts of patients with cardiomyopathy were utilised: (1) patients with cardiomyopathy seen at the specialist cardiomyopathy clinic at GSH, Cape Town between February 1, 1996 and December 31, 2009; and (2) a group of HCM patients and first degree relatives seen in a specialist cardiogenetic clinic at Tygerberg Hospital, who underwent CMR at GSH.

29 out of 109 unrelated cases with DCM had familial disease, and had a significantly younger mean age of onset of cardiomyopathy than non-familial cases. Two of the 29 patients with familial DCM

had at least one relative who was diagnosed with PPCM. Pedigree analysis of the 29 families was consistent with AD inheritance in 72.4%, AR inheritance in 17.2% and X-LR inheritance in 10.4%. Cardiac chambers were significantly more dilated with poorer LV systolic function in idiopathic than familial cases. The mortality rate of 40% after a median follow-up of 5 years was, however, similar in both groups. The presence of New York Heart Association functional class III and IV symptoms was an independent predictor of mortality, whilst heart transplantation was an independent predictor of survival in both groups. Digoxin use without serum monitoring was a significant predictor of mortality in idiopathic DCM.

We also presented the longest follow-up study of PPCM in Africans, showing (1) a case fatality rate of 16.7% over a median of 4.3 years in patients on modern medical therapy; (2) PPCM is associated with adverse outcomes; (3) chronic heart failure occurs in 80% of patients studied, and with virtually no normalisation of LV dysfunction; and (4) that persistence of symptoms of heart failure are the strongest predictor of death in patients with PPCM. We compared the time of onset of symptoms, clinical profile and outcome of patients with HHFP and PPCM and found onset of symptoms to be postpartum in all PPCM patients and antepartum in 85% of HHFP. PPCM was associated with more severe complications and a higher rate of mortality.

Clinical presentation of HCM in South Africa was similar to that reported in other studies from other parts of the world. The South African founder mutations that cause HCM were not found in the 42 probands studied at GSH and 29% of those tested for mutations in 15 genes had disease causing mutations in MYH7 and MYBPC3. Overall survival of HCM was 74% at 10 years, which was similar to the general South African population. The independent predictor of mortality was the presence of multiple genetic variants with or without a disease-causing mutation in any of the 15

genes tested. Finally, we investigated the correlation between genotype and CMR phenotype in HCM patients with the 3 founder South African mutations and found variable phenotypic expression associated with the different genotypes.

Indeed, the studies in this thesis have contributed to our evolving understanding of the complex disease concepts of pathophysiology, clinical presentation, natural history and outcomes in various cardiomyopathies in Africans. Future studies are needed to test the hypothesis that cardiomyopathies represent a continuum of the same disease spectrum with identical genetic abnormalities and disease onset, clinical and pathological features, disease progression and severity, and disease prognosis that is variable and is modulated by different host and environmental factors. In the future, larger, multicentre studies of the molecular genetic underpinnings of cardiomyopathy among Africans should lend greater support to this hypothesis.

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